## FINAL REPORT

## FIELD ECOLOGICAL ASSESSMENT

NATIONAL LEAD SITE PEDRICKTOWN, SALEM COUNTY, NJ

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## EXECUTIVE SUMMARY

A field ecological assessment was conducted at the National Lead Superfund Site in Pedricktown, New Jersey to gather empirical data for the evaluation of ecologically based lead (Pb) sediment and soil clean-up goals. This investigation is part of an integrated evaluation of the ecological concerns which exist at the National Lead Site.

This investigation consisted of solid phase toxicity testing, organism Pb accumulation studies and a habitat evaluation.

Fourteen-day solid phase toxicity testing of sediment using the midge Chironomus tentans revealed that survival chronic effects occurred at a sediment concentration of 1100 mg/kg (dry weight). Mortality occurred at the two lowest concentrations, however, the cause of the mortality appears to be related to depression of the water column pH, although alteration of Pb availability can not be discounted. Whole body analysis of green frogs (Rana clamitans) collected within the East and West Stream drainages showed Pb tissue levels ranging from 1.4 mg/kg to 23 mg/kg, with mean wet weight concentrations of 6 and 9 mg/kg, respectively. Frog tissue concentrations reflected sediment Pb concentration ranges.

In-situ earthworms (Eisenia foetida) exposures were conducted using a range of soil Pb concentrations (120-6,900 mg/kg dry weight). The maximum accumulation in the earthworms was 170 mg/kg Pb, over the 28-day exposure period. Earthworm tissue levels were related to soil organic matter (p=0.06) but not soil Pb concentrations. This suggests an interaction of soil Pb content and organic matter with lead availability. It was also found that the bioaccumulation factors (BAF, tissue concentration divided by soil concentration) decreased exponentially with increasing soil Pb concentration. Within the 28-day study some mortality of earthworms did occur, however, these deaths could not be attributed to Pb exposure.

White-footed mice (Peromyscus leucopus) were collected from areas representing three ranges of soil Pb contamination. Tissue Pb levels in the white-footed mice ranged from <1 to 13 mg/kg wet weight. Differences in whole body tissue Pb levels were found only between the area containing the lower Pb levels and the area containing the highest Pb levels.

#### 1.0 INTRODUCTION

## 1.1 Objectives

The objective of this study was to collect empirical data on target receptors and surrogate organisms to be used in a subsequent ecological risk assessment of lead (Pb) contamination surrounding the National Lead The results of this study and the Superfund site. ecological risk assessment will be used to evaluate ecologically based clean-up goals for soils and sediment The following specific areas of concern at the site. were addressed in this study: bioaccumulation of Pb by aquatic and terrestrial fauna exposed to contaminated sediment and soil, laboratory toxicity evaluation of sediment, and existing habitat evaluation.

## 1.2 Site Background

The NL site is located in Pedricktown, Salem County, New Jersey (Figure 1). It is an abandoned secondary lead smelting facility located approximately 1.5 miles east of the Delaware River. The facility is situated on 44 acres and was operated from 1972 to 1984. Lead batteries and other lead materials were handled by this facility<sup>(1)</sup>. Contaminants of concern include lead (Pb), cadmium (Cd), and chromium (Cr), however, the primary contaminant of concern is Pb.

During the treatment process at the NL Industries, battery acids were neutralized and lead battery plates were shredded. The pieces of lead were then fed into a rotary kiln where they were heated using coke or coal as a fuel source. It is believed that the waste coke/coal and other waste products from the kiln were dumped into slag piles still present on site. Contamination throughout the area surrounding the former facility is apparently largely the result of former airborne deposition from the smelter, and surface runoff into the streams.

The area of investigation for this study included two roughly parallel streams running along both the east and west sides of the former facility (referred to hereafter as the East and West Streams, respectively), and forested areas which are within their respective watersheds<sup>(1)</sup> (Figure 2). According to the Remedial Investigation (RI) surface Pb concentrations in soil range from 35 milligrams per kilogram (mg/kg) to above 20,200 mg/kg; sediment concentrations are approximately in the same range<sup>(1)</sup>.

### 2.0 METHODOLOGY

## 2.1 X-Ray Fluorometry (XRF) Procedures

XRF was used as a screening tool throughout this study to identify the range of Pb contamination in different areas of the site. A Spectrace 9000 portable XRF spectrometer was used in two different modes. Surface soil readings were taken using the instrument in-situ, by scraping leaves and other surface litter away from a 6by-6-inch area with a decontaminated stainless steel The XRF measurement probe was then placed directly on the prepared area for analysis. approach was applicable for situations when the soil was relatively dry and was used to characterize surface contamination in areas used for the earthworm and small mammal studies. For screening sediment samples, sediment was collected and homogenized in 5-gallon (gal) stainless steel buckets using decontaminated stainless trowels. Three aliquots were taken and oven dried before XRF readings were taken (see Section 2.2.3).

The instrument was regularly calibrated in the field by taking measurements on a set of standards. All XRF readings were recorded in a field logbook, entered into the instrument memory, and later downloaded onto a computer disk. All XRF procedures followed U.S. EPA/ERT SOP # 1713, Spectrace 9000 Field Portable X-Ray Fluorescence Operating Procedures. Detailed procedures and calibration data are provided in the Final Analytical Report - XRF Analysis (Appendix A).

## 2.2 Aquatic Study

## 2.2.1 Preliminary Fieldwork

Site reconnaissance was conducted on June 10 and 17, 1992, to search for a suitable reference location for the toxicity evaluation and to characterize water quality in order to establish testing protocol. Two water samples were collected from the East Stream and West Stream, both near Route 130. Each sample consisted of one, 1 liter (L) amber bottle for total organic carbon (TOC) analysis, and one, 1 L polypropylene bottle for hardness and alkalinity analysis. These were submitted to a subcontracting laboratory and results reviewed in order to establish protocol for the toxicity testing evaluation. Two minnow traps, baited with canned tuna, were set at the West Stream location on both days to identify potential target receptors of Pb contamination.

## 2.2.2 In-Situ Water Quality Measurement

Water quality measurements were recorded at three separate locations (Figure 2). These were at the West Stream near Route 130 (two locations), and the East Stream near Pedricktown Road. Measurements were taken during preliminary site visits, and during the sediment sampling event approximately seven weeks later. A Hydrolab Surveyor II monitoring instrument was used in-situ to measure following parameters: temperature, the dissolved oxygen, conductivity, oxidation-reduction potential, and salinity. It was precalibrated and operated according to the Hydrolab Corporation Owner's Manual (1985, Revision A), and according to U.S. ERT/REAC Standard Operating Procedures (SOP) Hydrolab Surveyor II, Water Quality #2139, Management Systems.

## 2.2.3 Sediment Sampling

In order to measure potential toxicity of sediment in both the East and West Streams, sediment was collected from five locations (Figure 3), each representing a different target Pb concentration for comparison. The target concentrations were as follows: <100 mg/kg; 200 mg/kg; 500 mg/kg; 1,000 mg/kg; and 2,000 mg/kg Pb. These locations were selected from a total of 17 locations screened for Pb using XRF. Sample locations were marked in the field with labeled 2-foot (ft) wooden stakes.

Sampling followed U.S. EPA ERT/REAC SOP #2016, Sediment Sampling. At each location, the sediment was collected using a decontaminated bucket auger or stainless steel trowel and placed into a 2.5gallon plastic bucket. The sample was thoroughly Each of the 17 samples were brought homogenized. to a central staging area and a pH measurement was taken using an electronic pH meter. Then the three 2 to 3 gram aliquots for XRF screening were taken the bucket. They were oven-dried individually labeled aluminum weighing boats and screened using XRF as described above and in Appendix A.

The five sediment samples were selected using the following criteria: if the sample XRF screening value was within approximately 25% of the target concentration value, the sample was retained. In cases where multiple samples were collected close to the target concentration range, the sample with

an XRF concentration closest to the target concentration was selected. If the sample did not approximate one of the target concentrations, it was discarded. An exception was made for the 2,000 mg/kg target concentration. A 2,000-mg/kg sample was not located with multiple sampling attempts, therefore, a 5,810-mg/kg sample (as measured by XRF) was substituted.

For the retained samples, sufficient sediment was placed into appropriate glass jars and submitted for the following analyses: toxicity testing, Pb, TOC, and grain size analyses.

## 2.2.4 Sediment Toxicity Evaluation

The five sediment samples (locations 2, 4, 5, 8, 9,) described above were sent to a subcontracting laboratory for a 14-day toxicity evaluation of lethal and sublethal (growth) endpoints using the midge Chironomus tentans. Survival was noted on a daily basis, and growth effects were evaluated by measuring weight and length before and after the exposure period.

The test conditions were established prior to the study and were based on results of preliminary analyses of surface water samples collected on site. These results included Hydrolab Surveyor II measurements, Hach kit testing, and analytical data on TOC. During the toxicity evaluation, physical parameters of toxicity testing water, including pH, alkalinity, and total hardness, were measured by the testing laboratory. A detailed description of test conditions and toxicity testing procedures is given in Appendix B, Sediment Toxicity Testing RFP and Report.

## 2.2.5 Sampling of Aquatic Vertebrates

Aquatic vertebrates were sampled in order to determine whole body burdens of Pb at different locations in the East and West Streams. During the study, six minnow traps were set for three days. Three of these were placed in the East Stream just north of Pedricktown Road and upstream of the site (Figure 4). Three additional minnow traps were deployed in the West Stream just south of Route 130 and downstream of the site.

While fish were caught at these locations, no species were represented at all locations (see Section 3.1.1). However, green frogs (Rana clamitans) appeared to be common and were the only species recorded consistently at all locations. They were chosen as a target species to measure tissue Pb concentrations because of their position in the food chain and their limited movement within the system and occurrence at locations with varying Pb levels.

Frogs were collected from the East and West Streams, both up and downstream from the site (Figure 4), using hand-held nets during morning and evening hours. Headlamps were used to locate frogs at night. A portable electroshocking device (Coffelt Electronics, Inc., Model BP-4) was used to augment trapping success in the East Stream.

## 2.2.6 Dissection and Tissue Processing

Following capture, frogs were placed individually-labeled Ziploc bags and assigned a unique specimen number. All captured individuals were maintained alive on wet ice until processing. After a sufficient number of individuals for analysis were collected, the frogs were transported back to the REAC bioassessment laboratory in a Individuals were euthanized and dissected immediately thereafter. Whole body weight (to the length gram) and (to the millimeter) were recorded prior to dissection. dissection procedure included removing the stomach contents and rinsing the stomach with distilled water before its replacement into the body chamber. The intestines were removed and discarded. procedures were followed to avoid influencing measurements of Pb uptake into body tissues by including undigested material.

After dissection, the individual frogs were frozen and then homogenized by combining the whole body tissue with dry ice in a tissue blender. The homogenization cup was decontaminated between samples using soap and potable water, a 10% nitric acid rinse, and a distilled water rinse. The frozen homogenate was transferred in individual 8-ounce jars to the REAC analytical laboratory for Pb and percent moisture analyses. One rinsate blank taken from the homogenization cup was submitted to the REAC laboratory for Pb analysis.

## 2.2.7 Statistical Analyses

Extensive statistical analyses were not conducted for the majority of the aquatic data collected due to small sample sizes and area of sampling utilized. Frog Pb concentrations were compared between eastern and western sides of the study area using a t-test on means. Also, the whole body wet weight of frogs was compared to Pb tissue levels using correlation analysis.

Data reported from Tables 8-10 from Volume I of the RI<sup>(1)</sup> were analyzed using correlation analysis to determine if Pb concentrations and pH in surface water were correlated. A copy of these data is provided in Appendix C of this report.

## 2.3 Earthworm In-Situ Bioaccumulation Study

## 2.3.1 Selection of Chamber Locations

XRF screening was conducted during three separate field visits. During the first visit on June 17, surface soils in two areas of the site were screened for Pb contamination using XRF at 26 locations. The first area encompassed the woods adjacent to the power line right of way located to the east of the Exxon facility, and south of the railroad tracks (Figure 5). The second area consisted of the woods adjacent to the West Stream which are located west of the former facility and south of the railroad tracks.

Fifteen surface XRF readings were taken in the first area (locations E1-E15). Ten of these (E1were taken along a transect north/south, with reading locations approximately 20 feet apart. The remaining five readings (E11-E15) were taken at a point approximately 100 feet east of this transect, in a box pattern, with one reading taken in the middle. The dimensions of "box" were approximately 20-ft-by-20-ft to assess the (Figure 5). This was done variability of surface soils within a limited area. Eleven XRF readings were also taken along a north/south transect running through the second area (locations W1-W11).

Two weeks later, 32 additional locations throughout the eastern and western sides of the study area were screened. These were primarily used to determine the locations for the *in-situ* earthworm

bioaccumulation chambers. Ten of these readings were of an exploratory nature and were not mapped. These were logged on the instrument as locations E19-23, E25, and W26-29. The locations of the remaining readings (E16-E18, E24, E26-E29, W12-W25) are shown in Figure 5.

Twenty locations (Figure 7) were selected in order to measure bioaccumulation across a range of target concentrations. The range of concentrations was established prior to the study and was based upon results of the initial XRF Target concentrations ranged from 100 survey. mg/kg to 10,000 mg/kg, as identified in the REAC Quality Assurance Work Plan (QAWP). Difficultly was experienced in locating the target Pb levels in field, due to the variability in soil concentrations with depth. Thus it was often necessary to homogenize soil with varying Pb concentrations to achieve the target concentration level. As a result, the ultimate concentrations to which the worms were exposed reflect the target concentrations, and not necessarily the surface soil concentrations at the chamber locations. Likewise, it is important to note that the soil exposures are not necessarily representative of actual exposure in the wild populations as field populations are exposed to varying Pb levels with soil depth and lateral movement in the soil.

## 2.3.2 Chamber Installation and Soil Sampling

Eisenia foetida were used to test for in-situ bioaccumulation of Pb over a 28-day period. This species has been commonly used to test organism in the literature and has been successfully used in other in-situ studies. Earthworms were obtained from a clean stock culture obtained from the U.S. EPA Environmental Research Lab in Corvallis, Worms were maintained in a peat moss 4-7° C in medium at the REAC bioassessment laboratory prior to commencement of the study.

In-situ earthworm bioaccumulation chambers (Figure 6) were preassembled at the REAC facility. Each consisted of a 2 1/2-gal bucket with the central portion of the bottom removed. This was replaced by fine nylon netting and an overlying piece of 1/4" stainless steel hardware cloth. The central portion of the bucket lid was also removed, leaving a 1" margin around the rim. A second piece of nylon netting was placed across the top of the

bucket and held in place by closing the lid. This allowed precipitation to enter the chamber and provided adequate ventilation.

Test chambers were installed from June 25-26, 1992. The protocol for chamber installation was developed prior to the study. Personnel worked in two-person teams, with each team assigned to individual test locations.

At each location a hole was dug just wide and deep enough (about 14" in diameter and 12" deep) to implant the plastic chamber such that the lid of the chamber was even with the surface of the ground. This allowed the screened lid to remain open for aeration. Once the hole was dug, a test chamber was placed into each hole, and the dimensions of each hole was fine tuned to fit the container.

As soil was removed from each hole it was passed through a sieve constructed from a 3/8 " stainless steel hardware cloth. During the sieving process soil was collected into a large stainless steel tray or onto a large sheet of polypropylene plastic. Rocks and larger roots were removed. sieved soil was then homogenized by mixing it into a cone-shaped pile using a stainless steel trowel. Subsequently, an in-situ XRF reading was taken from the sieved, homogenized sample. It was found that Pb concentrations were significantly higher at the ground surface than below making it difficult to obtain target concentrations. As a result, it was often necessary to homogenize a greater proportion of surface soil into the pile to achieve a value close to the target concentration. In many cases, was also necessary to take multiple until the target concentration readings, received.

Once the target concentration was received, aliquots were then collected into glass jars for each of the following analyses: atomic absorption spectrometry (AA) analysis of Pb (four-ounce jar), TOC (four-ounce jar), and grain size (32-ounce jar). Glass jars were prelabeled with a REAC sample number, and the sampling location was written in with a permanent marker immediately after taking the sample.

An electronic soil moisture meter was inserted into the pile of homogenate to determine initial soil moisture. A pH meter was used to check soil pH at each location by preparing a slurry with approximately 5 g of soil and distilled water. The slurry was mixed in a corner of the stainless steel tray or plastic sheet, and the pH probe was inserted into the slurry.

After Field measurements were taken and the soil samples were collected, the remaining soil was used to fill the earthworm chamber to within 1" of the top. This 1" gap allowed air exchange and gave the worms the opportunity to migrate to the surface of the chamber at night. Additional soil was packed around the sides of the chamber to secure it.

Two additional chambers were installed within a 1-ft radius of chamber 4. This was done in order to check earthworm mortality after 10 and 20 days into the study, respectively. The objective of this mortality check was to insure that mortality was not occurring due to soil moisture, lack of organic material, or extreme fluctuations in temperature or precipitation. Location 4 was chosen because a suitable location in a similar habitat that was free of Pb contamination could not be found in the immediate area. The Pb concentration of the soil homogenate at this location was 180 mg/kg.

All equipment used for soil sampling and chamber installation (shovels, trowels, sieves, etc.) were decontaminated between locations. Decontamination followed the sequence below:

- o Soil and other material were physically removed.
- Equipment was washed with soap and potable water.
- Equipment was rinsed with distilled water.
- Equipment was sprayed with 10% nitric acid.
- Equipment was rinsed again with distilled water.
- Equipment was allowed to air dry.

Sample handling and documentation followed U.S. EPA ERT/REAC SOPs #2002, Sample Documentation, and 2003, Sample Storage, Preservation and Handling

## 2.3.3 Test Conditions and Chamber Monitoring

After all chambers were installed, the test worms were removed from their culture media in random lots of ten worms, rinsed with distilled water, and

weighed to the nearest 0.001 g at a central location. Individual weights of test worms were recorded in groups of ten. This allowed the mean worm weight per chamber to be calculated both before and after the 28-day bioaccumulation period. Worms were then placed within moistened filter paper into sealed Ziploc bags with small air holes at the top. Each bag was randomly labeled with a chamber location number from 1 to 20 and placed in a cooler with wet ice to maintain low metabolic rates. Subsequently, the worms were transferred by cooler from the central processing area to the field.

On June 26, ten Eisenia were placed on top of the soil in each chamber. The nylon netting and lid were then placed over them and the lid sealed. The soil surrounding the chamber was packed around the lid to help discourage potential predators. Five randomly chosen chambers were checked following installation to ensure that the worms had burrowed into the soil matrix.

Near the onset of the study, an additional worm sample (ten worms) was taken from the source laboratory population and analyzed to determine the Pb concentration present in the test worms prior to their exposure to on-site soils. The depuration and tissue homogenization procedures followed were identical to those described below for the test worms.

To monitor soil temperature throughout the study, a TempMentor (TM) recording soil thermometer was installed at chamber location 4 to record temperature over the 28-day period. The datalogger was preset to record temperature readings on an hourly basis throughout the study. Results were downloaded onto a portable computer at the end of the study. A rain gauge was installed at the same location to measure precipitation.

The two additional chambers at location 4 were checked at 10 and 20 days respectively. Recovered worms were rinsed with distilled water, inspected for lesions, depurated, and then submitted to the REAC analytical laboratory for Pb and percent moisture analyses.

At the end of the 28-day period, all worms were removed from the 20 test chambers. Each chamber was removed from the ground by gently pulling on

the handle. Soil was gradually poured from the chamber onto a clean plastic sheet. The soil was gently sorted by hand and earthworms were set aside on the sheet until no additional earthworms could be found in the chamber. In cases where ten earthworms were not found, the soil in the chamber was completely rechecked two additional times. Earthworms were rinsed in the field with distilled water and inspected for lesions, then placed into depuration chambers labeled by chamber location. The depuration chambers consisted of plastic containers with moistened filter paper at the bottom. The depuration chambers were placed on wet ice in a cooler for transport back to the REAC facility.

## 2.3.4 Tissue Processing

At the REAC facility, the depuration chambers were removed from the cooler. Since worms were initially weighed with soil in them, the final weights were recorded by chamber before depuration. Subsequently, all worms were depurated at 75° F in the chambers. An earthworm was considered fully depurated if no soil was visible in its alimentary tract when it was placed onto a light table. earthworms were fully depurated after 72 hours. Then the worms were homogenized with dry ice and placed in a 40-ml vial labeled with the chamber number. The frozen homogenate samples were submitted to the REAC analytical laboratory for Pb and percent moisture analyses.

## 2.3.5 Statistical Analyses

Regression analysis was used to compare the analytical results of Pb concentrations in earthworm tissue with corresponding Pb concentrations in the soil samples taken from each All comparisons were made using dry chamber. weight concentrations of earthworms and AA data on Pb concentrations. Since soil concentration is not necessarily the only variable affecting Pb uptake by earthworms, the influence of other variables such as TOC, grain size, and pH was also investigated using a step-wise regression model. Data on Pb soil concentrations, TOC, pH, and grain size of soil collected from each location were entered into this model as predictors of earthworm Pb tissue concentrations.

Bioaccumulation factors (BAFs) were calculated at several different soil concentration ranges to measure the proportion of Pb absorbed into earthworm tissue. These were calculated as:

BAF = Pb concentration in earthworm tissue + Pb concentration in soil.

In the REAC QAWP, it was stated that data would be input into an equilibrium accumulation model for earthworms developed by Connell and Markhum<sup>(2)</sup>. However, this model was developed exclusively for volatile organic compounds (VOCs), and upon further investigation could not be adapted for use in predicting Pb uptake.

## 2.4 Small Mammal Study

## 2.4.1 Field Trapping Design

The white-footed mouse (Peromyscus leucopus) was identified as a target species on the basis of habitat availability noted during preliminary field surveys.

Small mammal trapping was conducted in three discrete wooded areas of the site identified during the preliminary field visit (Figure 8). These areas were chosen upon the basis of available small mammal habitat, prior extent of contamination studies<sup>(1)</sup>, and preliminary XRF field screening. The three grid areas of the site were in the following locations. Grids I and IA were in the woods west of and adjacent to the former facility, and immediately south and north of the railroad tracks, respectively. Grid II was in the woods located to the east of the former Exxon facility, and south of the railroad tracks. Grid III was in the woods located about 400 ft south of Grid II (Figure 8).

In this report, Area I refers to the area encompassing both the Grid I and Grid IA areas. This area is dominated by red maple (Acer rubrum) with some arrowwood in the understory. While this forested area is primarily associated with the West Stream, it contains some upland areas as well. The area immediately adjacent to the stream is open and shrubby, dominated by arrow-leaved tearthumb

(Polygonum sagittatum) and elderberry (Sambucus canadensis). Anticipated surface concentrations of Pb in this area, based upon preliminary screening, were approximately 2,000 mg/kg.

Area II encompasses the immediate vicinity of Grid II, and is dominated by sweetqum (Liquidambar styraciflua), some black and cherry (Prunus Occasional gaps in the canopy allow serotina). scattered ground cover of arrowwood viburnum (Viburnum dentatum), wild blackberry (Rubus sp.), wild currant (Ribes sp.). Surface Pb concentrations throughout most of this area were anticipated to be in the range of 500-1000 mg/kg based upon initial screening.

Area III consists primarily of forested wetland, with upland deciduous forest along its southern periphery. Grid III is encompassed within this area. Most of this area was dominated by mature sweetgum trees. Although this area was not investigated using XRF during the preliminary field visits due to time constraints, Pb concentrations were anticipated to be in the range of 100 to 500 mg/kg based on stream sediment data collected in the RI<sup>(1)</sup>.

Small mammal trapping and additional XRF screening of surface soils were conducted from August 25 through 28. Ten lines of 15 traps each (100 Museum Special snap traps and 50 Sherman box traps) were set in each of three grids (I,II and III) for a total of 150 traps per grid (Figure 8). Museum Special lines were alternated with Sherman trap lines over the first ten lines set in each grid. The remaining five lines (lines 11-15) consisted entirely of Museum Special snap traps. Museum Special traps were baited with a mixture of peanut butter and rolled oats. Sherman traps were baited with rolled oats.

Due to the low trap success for Grid I (<1.0%) on the first evening, further XRF screening was conducted in an adjacent area, to determine if the number of traps at this Pb concentration range could be expanded. An alternate area was located north of the railroad tracks. A total of ten readings were taken within this area. Five XRF readings (N1-N5) were taken along a north/south transect which began in the woods approximately 75 feet north of the railroad tracks, and extended northward 100 feet. Readings were taken approximately 25 ft apart in the same manner as previously described.

A second transect of five readings (N6-N10) was run perpendicular to the first. This extended for approximately 100 feet. These readings were also taken at 25-ft intervals. Subsequently, five additional surface XRF readings were taken along a transect running 50 feet north and parallel to the latter transect.

This area exhibited a concentration range (close to 2000 ppm Pb) similar to the Grid-I area. It also represented similar habitat and was separated only by railroad tracks. Thus, it was selected as an additional trapping area. A grid labeled IA (Figure 8) was installed in this area in order to augment trap success from the Grid-I area. One hundred traps were set and placed in five lines consisting of ten Museum Specials and ten Sherman traps.

Additional XRF readings were taken in Area III to characterize surface Pb concentrations. Two perpendicular transects were run through the area, with five XRF readings taken along each one. Points were labeled EE1-EE10 (Figure 5). Upon this further characterization of Area III, it was determined that the area had a contamination range of 438 to 4930 mg/kg; rather than the anticipated 100 to 500 mg/kg range.

All field trapping activities were conducted in accordance with U.S. ERT/REAC Draft SOP #2029, Small Mammal Sampling (Appendix D). However, a non-contaminated reference area was not identified within the immediate vicinity of the site for comparison to the site results.

## 2.4.2 Specimen Collection

Traps were checked twice daily during early morning and early evening hours and were reset until a target of ten *Peromyscus* was reached for each area. Traps were set for two nights in Grids I and III. Grid IA was trapped for one night, and Grid II was trapped for three nights.

Non-target species were released if live-trapped, and archived if caught in Museum Special traps. Peromyscus that were captured live were sacrificed

at the REAC bioassessment laboratory by cervical dislocation. All specimens were labeled with aluminum tags in the field, placed in coolers on site, and transported to the REAC bioassessment laboratory on a daily basis for processing.

## 2.4.3 Processing and Analyses

mammals were dissected and processed according to the U.S. EPA ERT/REAC Draft SOP for Small Mammal Tissue Dissection and Processing (Appendix D). This included taking body and organ metrics, recording the species, age and sex of specimens, and collecting a small section of tissue from both the liver and kidney for analysis of histopathological effects. The histopathology samples were preserved in glass vials filled with 4 percent (%) buffered formaldehyde solution.

The contents of the gastrointestinal (GI) tract were removed from each specimen. The intestines were discarded. The stomach was subsequently rinsed with distilled water before returning to the body cavity. A whole body tissue homogenate was then prepared and submitted for analysis to the REAC central laboratory for analysis of whole body Pb content, and percent moisture.

## 2.4.4 Statistical Analyses

Comparison of metric data between areas poses a statistical problem, since organ weights or other metrics may covary with body weight. According to Greene (3) standardization of such data into ratios dividing by body weight can result heterogeneity of variance, biased estimates of the distributions, mean, non-normal and other statistical intractable problems. recommended that Analysis of Covariance (ANCOVA) be followed in such cases. Organ metrics which were normally distributed were thus subjected to the criteria below to determine whether Analysis of Variance (ANOVA) or ANCOVA were appropriate for analysis. The criteria for test selection and the subsequent analyses followed the suggested approach listed in the SAS Institute, Inc. Anova and regression course notes (4).

All metrics data (total weight and organ weights) were first tested for normality. For data which were normally distributed (total weight, adrenal weight), all slope parameters were tested to

determine whether or not they were equal to 0. The slope parameters did not differ significantly from 0 for either variable, so comparisons between areas were made using ANOVA.

For the remainder of the data set, the individual regression lines of body weight (y) versus organ weight (x) were compared between trapping areas to determine if the lines were equal. If there were no significant differences between slopes, then ANCOVA was used to determine if statistically significant differences occurred between areas. This was the case for liver and kidney weights. Spleen, thymus, and uterus data showed significant differences between slopes. These data were analyzed using a non-parametric test (Kruskal-Wallis Test<sup>(5)</sup>).

Data on Pb concentrations in mice were also compared by area using an ANOVA model. Data were first converted to dry weight due to individual differences in percent moisture. Then they were transformed using a log transformation, since data were not normally distributed. Pb concentrations in mice were also compared with body weight using correlation analysis.

#### 2.5 Terrestrial and Wetland Habitat Assessment

During the above field investigations, dominant vegetative cover types were noted, and subsequently a vegetation map of the site area was prepared. This included wetland habitats, although wetlands were not delineated, because a wetland delineation was conducted during the prior study<sup>(1)</sup>. A species list was developed for plants and vertebrate animals which were observed on site.

## 3.0 RESULTS

## 3.1 Aquatic Study

## 3.1.1 Preliminary Fieldwork

A water sample collected from the East Stream near Route 130 exhibited a hardness value of 62.5 milligrams/liter (mg/L) and an alkalinity value of 100 mg/L. The pH value was measured at 7.5, and total dissolved solids were measured at 22 mg/L. The TOC analysis revealed a concentration of 18 mg/L (Appendix E - Analytical Report).

A water sample collected from the West Stream near Route 130 revealed a hardness value of 47.5 mg/L and an alkalinity value of 15 mg/L. The pH value was 6.6 units, and the TOC analysis revealed a concentration of 13 mg/L. These results were used to establish toxicity evaluation protocols for the 14-day test using Chironomus tentans (see RFP-Appendix B).

Two minnow traps set in the West Stream near Route 130 revealed that red-breasted sunfish (Lepomis auritus) and crayfish (Orconectes sp.) were potential target receptors for investigation. However, it was subsequently determined that of the areas sampled this was the only location in which they were found.

## 3.1.2 In-Situ Water Quality Measurement

Temperature readings taken with the Hydrolab Surveyor II in the West Stream location near Route 130 were higher than readings taken at the East Stream location near Pedricktown Road (Table 1). The water temperature reading taken on June 17 at 1340 hours at the East Stream was 21.3° C. Twenty minutes later, at the West Stream location the temperature was 6° C higher. On August 10, the temperature was 2° C higher at the West Steam location than at the East Stream.

The pH measurements were fairly consistent, varying from 5.2 to 6.5 over all locations, throughout the study (Table 1).

Strong differences in dissolved oxygen (DO) were noted between locations, and between sampling dates in June and August. Readings taken on June 17 were similar to locations within the East and West stream, at about 6 mg/L. However on August 10, the DO values observed in the East Stream (Pedricktown Rd) and West Stream (Route 130) differed by more than 8 mg/L. Moreover, a positive difference in DO values was observed between June 17 and August 10 at the West Stream, while they sharply decreased at the East Stream location when the two dates were compared.

Conductivity was fairly constant between locations throughout the study, with the exception of a decrease observed in the East Stream between June 17 and August 10 (Table 1). Oxidation-reduction

potential varied 0.044 to 0.183 volts throughout the study, and salinity was consistently measured at 0.00 parts per thousand (ppt).

## 3.1.3 Sediment Characteristics

XRF screening revealed Pb concentrations were generally lower in sediment samples collected within the East Stream than the West Stream drainage, and were below 1,400 mg/kg in all samples (Table 2). Concentrations varied widely in the West Stream drainage, ranging from 386 mg/kg to over 20,000 mg/kg, as measured by XRF.

The highest concentrations were recorded in the area of the West Stream from the railroad tracks south to Pedricktown Road (4,147-20,670 mg/kg), represented by sampling locations 8,11, and 13 (Figure 3). Data are summarized in Table 2. Raw XRF data for each sampling location is given in Appendix A - Table 1. Sediment samples are listed with the letters B, C or D following the sample number. These letters represent the specific aliquot analyzed, since three aliquots per sample were analyzed using XRF.

The Pb concentrations of sediment samples chosen for toxicity testing (Table 3) came close to approximating the target concentrations, with the exception of the 2000 mg/kg target concentration.

All five locations had similar sediment pH values (near 6) with the exception of location 2, which had a pH reading of 3.9.

TOC values in sediment ranged from 4 to 160 g/kg. Pb concentrations did not appear to be directly related to TOC content in the sediment.

Grain size analysis of sediment revealed that the proportion of fine particles (silt and clay) in the five samples ranged from less than 5% to greater than 75% (Table 4). Location 5 had a noticeably lower silt and clay component, and consisted of approximately 85% sand and fine sand. This sample also had the lowest TOC concentration of any location analyzed. The remainder of the samples consisted of greater than 50% fine sand, silt and clay.

## 3.1.4 Sediment Toxicity Evaluation

A 14-day solid phase toxicity test using the midge Chironomus tentans revealed statistically significant differences in survival among treatment groups exposed to Pb-contaminated sediment from different locations (Table 5 and Appendix B). Mortality did occur within the test but appears to be associated with a depressed alkalinity and pH in the water column rather than directly related to the Pb level in the sediment. Survival was high (86.7%) not only in the laboratory control group, but also in groups exposed to sediment collected at locations 4 and 8, where Pb concentrations were high (1100 mg/kg and 4400 mg/kg, respectively). Conversely, survival was low in the three groups where sediment contamination was low (53, 260, and 670 mg/kg Pb, respectively).

Growth parameters could not be measured in three of the samples due to mortality. At sample locations 4 and 8, C. tentans there was not mortality relative to the control, however, length and weight measurements demonstrated significant differences in growth relative to the control. Mean length of midge larvae exposed to sediment from location 4 was 15.32 mm, which was significantly less than the mean length of control larvae (18.80 mm) at the conclusion of the test (ANOVA, F=4.26, 11 d.f., Dunnett's Test, p<0.05) (Appendix B, p.34). There was no significant difference in length between the control group, and midges exposed to sediment from location 8, which had the highest sediment Pb concentration used in the test.

The mean dry weight for midge larvae exposed to sediment from locations 4 and 8 (0.00148 g, and 0.00166 g, respectively) was significantly less than that of the control mean (0.0311) at the end of the 14-day test (ANOVA, F=4.26, 11 d.f., Dunnett's Test, p<0.05, Appendix B, p.39).

Measurements of physiochemical characteristics of the water used in the toxicity evaluation showed major changes in the alkalinity and pH in the tests utilizing sediment from locations 2, 5, and 9 (Table 6). In these three tests the water column alkalinity was reduced to less than 12 mg/L, while in the two remaining samples and control, alkalinity ranged from 16-56 mg/L. Similarly, pH dropped below 4.5 units at some point during the study in the exposures for sediments 2, 5 and 9,

while not the other exposures. The dilution water used for all tests was the same and was initially 20 mg/L. Hardness measurements indicate that no significant alteration in water hardness occurred during the test.

## 3.1.5 Pb Accumulation in Green Frogs

A total of 13 green frogs were caught on site during a two-day period in August. Four green frogs were caught in the East Stream upstream of the site. Five were caught in the West Stream just south of the railroad tracks (Figure 4). Two were caught in a small (approximately 20-ft-long-by-10-ft-wide) pool in the West Stream just north of Pedricktown Road. Two additional frogs were caught in the West Stream north of the railroad tracks and south of Route 130. While catching the green frogs, one bull frog (Rana catesbeiana) was caught and released.

Whole body (wet weight) Pb concentrations of green frogs ranged from 1.4 to 23.0 mg/kg over 13 locations within the East and West Stream drainages (Table 7 and Appendix E). The five frogs captured in the East Stream near Pedricktown Road, and in the West Stream near Route 130 had lower whole body Pb levels than the frogs captured in the West Stream south of the railroad tracks (Table 8).

The mean wet weight Pb concentration of the four frogs caught in the East Stream drainage was 5.98 versus 9.20 mg/kg for the nine West Stream frogs, however, these differences were not statistically significant (Appendix G).

No relationship between fresh weight and whole body wet weight Pb concentrations was evident (Table 6). Percent moisture values were fairly consistent, ranging from 74 to 86% of fresh body weight.

Body length (vent to snout) of captured frogs varied from 30-60 mm. Total fresh weight varied from 1.889 to 15.988 g (Appendix F).

## 3.2 Earthworm In-Situ Bioaccumulation Study

## 3.2.1 Soil Characteristics

## 3.2.1.1 Surface Pb Concentrations (XRF)

Surface Pb concentrations (Table 9 and Appendix A) measured using in-situ XRF were notably higher than concentrations a few inches below the surface. This trend was noted during preliminary screening for selection of the earthworm chamber locations. At several locations (such as E1, W8, W11, W13, and W22) where in-situ readings were taken at multiple depths, surface concentrations of Pb were substantially higher than below the surface (Appendix A - Table 1).

For example, at location E1 the XRF reading was 3041 mg/kg Pb at the surface, and 1537 mg/kg Pb at 6" below ground surface (bgs). At location W22, the XRF reading was 5394 mg/kg Pb at the surface and 2807 mg/kg at a 3" bgs.

At locations W8 and W13, the XRF readings taken from the surface humus layer were about one and a half times the values recorded at 1" bgs. At location W8, the XRF reading was 2048 mg/kg Pb at the surface, and 707 mg/kg at 1" bgs. At W13 the XRF reading was 2902 mg/kg Pb at the surface, and 737 mg/kg Pb at less <1" bgs (Appendix A - Table 1).

Surface Pb concentrations also varied considerably within a relatively small radius. For example, XRF readings at points E11-E15 were taken within a 25-ft-by-25-ft square-shaped area. Four readings were taken at each corner, and one reading was taken in the center (Figure 5). Within this area, surface Pb concentrations ranged from 169 to 902 mg/kg.

At two other locations, replicate XRF readings were taken at a distance of 2-feet away from At point E19, the the original reading. initial reading was 1501 mg/kg Pb, while two away a reading of 1146 mg/kg was feet The variability is believed not to recorded. be a function of measurement variability. test measurement variability at location E29, a surface reading of 3979 mg/kg Pb was recorded, with a duplicate reading of 3958 mg/kg at the same exact point. However, a value of 333 mg/kg Pb was recorded at a distance of 2-ft away.

## 3.2.1.2 Chamber Pb Concentrations (XRF and AA)

Pb concentrations of soil homogenate as measured by AA were lower than concentrations measured by XRF at most of the chamber locations (Table 9, and Appendix E). values as measured by AA in soil homogenate used to fill the earthworm chambers ranged from a low value of 120 mg/kg at location 20 to a high value of 6900 mg/kg at location 18. XRF readings from the same set of samples gave a range of concentrations from 169 mg/kg to 10,082 mg/kg Pb (Table 9). XRF screening indicated location 4, with a value of 169 mg/kg Pb, had the lowest concentration of the 20 locations. AA analysis confirmed that location 20 was actually lowest in Pb content, yielding a value of 120 mg/kg.

Despite the above differences, XRF and AA values corresponded well. A regression analysis which used Pb concentration measured by AA as the dependent variable and Pb concentration measured by XRF as the independent variable yielded an r<sup>2</sup> value of 0.985. The regression was highly significant (F=1269.6, 19 d.f., p<0.0001). This model was run without an intercept.

# 3.2.1.3 pH, Percent Moisture, and Total Organic Content (TOC) in Chambers

Soil pH values were fairly consistent between chambers, being close to neutral (Table 10). Sample location 9 was somewhat lower, with a pH value of 6.2 units. All sample locations east of the Exxon facility (Figure 7) had a pH value of 7.0 units.

Soil moisture in the earthworm chambers measured in the field with a moisture meter varied from 5 to 68%. Samples on the east side of the facility were notably drier, as locations 1-7 ranged from 5 to 20% moisture. Sample location 20, taken from the edge of the East Stream, had 55% moisture. Sample locations on the western side of the facility (8-19) exhibited variable moisture content (Table 10).

Soil moisture values measured by the subcontracting laboratory (Table 10) revealed

a similar overall trend, but in several cases (locations 3, 5, 6, 10-12, and 14) considerable differences between field and laboratory measurements were noted. For example, at location 14 the difference between field and laboratory measured percent moisture was about 40%.

Soil Pb levels in the earthworm chambers were correlated with TOC concentrations (r=0.35, n=20). TOC of the chambers installed on the east side of the facility were significantly lower than that of those on the west side of the facility when chamber 20, the wetland reference chamber, is omitted (t-test for unequal variance, 15.7 d.f., p<0.036). The mean TOC of chambers on the eastern side (locations 1-7) was 39 grams per kilogram (q/kq), versus a mean value of 124 g/kg for chambers on the western side of the facility.

## 3.2.1.4 Grain Size Characteristics of Chamber Soil

Results of grain size analysis of soils collected from earthworm chambers show no major differences in grain size between the eastern (locations 1-7) and western areas of the site. Results indicated that soils consisted primarily of sand and other large particles (Table 11). With the exception of location 16, which contained 16.26% fine particles, soil samples collected throughout area contained less than 12% particles (silt and clay). Soil concentrations (log-transformed) were highly correlated with the silt/clay percentage of the soil in the western chambers (r=0.95, n=20) (Appendix E).

A significant proportion of the samples submitted for grain size analysis consisted of vegetative material and organic detritus, which was measured by the percentage weight of the sample burned off immediately prior to the grain size analysis (Table 12, and Appendix E). At several locations, the proportion of organic matter was greater than half of the sample, and the samples collected at locations 14 and 15 had organic matter in excess of 75%. The grain size analyses were conducted on the remaining portion.

Percent organic matter ranged from 4-22% in chambers in the eastern side of the facility (locations 1-7). With the exception of location 16 (17.87%) organic matter ranged from 30-70% in chambers on the western side. Soil in the eastern chambers, with a mean of about 13%, contained a significantly lower percentage of organic material than soil in the chambers, which had a mean close to 47% (T-test for unequal variance, p<0.001, 15.7 d.f.). Location 20 was not considered in this analysis, since it was a wetland reference sample located in the eastern area with soil similar to that on the west side.

## 3.2.2 Environmental Data

Soil data measured by the TempMentor<sup>(TM)</sup> thermometer on an hourly basis at location 4 revealed regular diurnal fluctuations in soil temperature of up to 6°C (Appendix H). Soil temperature was generally lowest between 0600 and 0800 hours, and was consistently highest at approximately 1800 hours (Appendix H).

Soil temperature generally increased throughout the 28-day exposure period (Appendix H). The lowest temperature (16.8° C) was recorded on June 29 at 0800 hours. The highest temperature (25.9° C) was recorded on July 20 at 1700 hours.

A total of 2 3/16" rainfall was recorded in the rain gauge at location 4 during the 28-day exposure period. Most of this precipitation (1 1/4") fell during the last ten days of the study.

## 3.2.3 Earthworm Survival and Pb Accumulation

## 3.2.3.1 Survival

Worm survival equalled or exceeded 70% in all chambers except at locations 11, 12, 16, and 18-20 (Table 13). No worms survived in chambers at locations 11, 12, and 19. Cursory inspection of the data reveals that mortality was not correlated with Pb concentrations. Worms at location 20 were exposed to only 120 mg/kg Pb yet exhibited high mortality (70%). Conversely, worms at location 17 which were exposed to 6800 mg/kg exhibited far lower (30%) mortality. Nine of the 10 worms in the ten-day exposure group exposed to on-site soil

at location 4 were retrieved. In the 20-day exposure group at the same location, all ten worms were retrieved.

When checking chambers in the field at ten day intervals, some mortality was noted, in that dead worms were found at the top of the chamber soil. However, in some chambers where there was no trace of mortality during these checks, such as at location 20, chamber mortality was significant.

## 3.2.3.2 Pb Accumulation in Earthworms

No Pb was detected in the laboratory reference worms at a detection limit of 0.25 mg/kg. The test worms exposed to 180 mg/kg Pb soil at location 4 contained 15 mg/kg Pb (wet weight) after ten days and 21 mg/kg Pb (weight) after 20 days (Table 13).

Earthworms exposed to Pb contaminated soil for 28 days accumulated Pb ranging from 29 mg/kg to 170 mg/kg Pb (Table 13). Pb concentrations of earthworms (dry weight, corrected for percent moisture differences between samples) were not correlated with soil Pb levels in the chambers (r=0.18, n=20). The wet weight Pb concentration of worms exposed to 6900 mg/kg, 2200 mg/kg, and 450 mg/kg Pb in soil was actually quite similar. Corresponding tissue concentrations were 140 mg/kg, 170 mg/kg, 130 mg/kg, respectively.

Bioaccumulation factors (BAFs) defined as the Pb concentration in worms divided by Pb concentration in soil, ranged from 0.01 to 0.29. BCFs varied with the soil concentration (Table 14). At lower concentrations, worms accumulated a higher proportion of Pb than worms at higher concentrations (Table 14). Worms exposed to Pb concentrations less than 500 mg/kg had a BCF of 0.23, while worms exposed to >6500 mg/kg Pb had a BCF of 0.01. plot of BCF values versus soil exponential concentrations suggests an relationship (Appendix I), with the proportion of Pb absorbed decreasing at higher soil concentrations.

Statistical evaluation of the soil parameters with Pb accumulation data confirmed that there

is not a simple relationship between earthworm accumulation and soil Pb concentration. Of the soil parameters measured only percent organic matter was statistically associated with Pb accumulation at the p=0.10 level (p<0.062), however a regression analysis shows that the relationship is weak with an  $r^2$  value of 0.23 (Appendix G).

Despite differences in organic matter in the chamber soils described above, earthworm Pb concentrations between the eastern and western sides of the facility were not significantly different (t-test for unequal variance, p<0.25, 10.4 d.f.).

## 3.2.4 Weight Changes in 28-day Exposure Worms

Initial mean wet weights for the ten worms placed into each chamber ranged from a low value of 0.440 g at location 13 to a high value of 0.670 g at location 9 (Table 15). Mean weights at 28 days ranged from 0.155 at location 18 to 0.375 at location 3. A pairwise comparison of mean wet weight of test worms before and after the 28-day exposure period corroborated that mean weight decreased significantly (T-test, T=11.81, 17 d.f., p<0.0001). Percent moisture in earthworm tissue was fairly consistent, ranging from 81-86% (Table 13).

Although the test worms in every chamber lost weight (Table 15), the magnitude of weight loss was positively correlated with the Pb content of the soil to which they were exposed (r=0.67, n=17). While the regression equation "weight difference = soil Pb content" was highly significant (F=27.34, 17 d.f., p<0.0001), the r<sup>2</sup> value was relatively low, at 0.44. Mean weight loss was highest at chamber location 18, where the soil Pb content was highest (Table 15).

## 3.2.5 Physical Condition of 28-day Exposure Worms

Most of the worms exposed to contaminated soils showed no overt signs of lesions or other abnormalities. However, a number of worms from varying Pb exposures did have physical abnormalities.

At location 4, of the ten worms recovered after 20 days, one had a notable lesion, and a second had a yellow-green pus-like substance near the clitellum.

Of the seven worms surviving at location 9, one had a prominent constriction immediately anterior to the clitellum. A similar constriction was noted on one of the seven worms surviving at location 17, and another worm had been severed in half.

At location 20, where only three worms were retrieved, all were disfigured or had a portion of their bodies missing. One had a prominent chunk of tissue missing on its posterior end, about 2/3 of the way below the clitellum. Another worm had apparently been severed just below the clitellum, while a third was totally disfigured but still alive.

## 3.3 Small Mammal Study

## Surface Pb Concentrations

XRF readings taken within the immediate vicinity (within 100 ft) of the small mammal grids revealed variability considerable in surface concentrations (Table 16). Inspection of the mean and median Pb concentration values for each of the three general areas investigated reveals that Areas I and II had surface Pb levels close to their anticipated concentration ranges of 1000-2000 mg/kg, and 500-1000 mg/kg, respectively. However, Pb concentrations Area III surface in significantly higher than the anticipated concentration range of <500 mg/kg.

The range of surface Pb concentrations encountered (Table 16) was considerable within each of the three areas measured. Area I had a mean surface Pb level of approximately 1650 mg/kg Pb, but the range of concentrations encountered was from 150 to 4200 Similarly, Area III had a mean of 2300 mg/kg Pb, with a range of 400 to 5000 mg/kg Pb.

#### 3.3.2 Trap Success

Overall trap success was less than 5%, for the 1,150 trap-nights in this study (Table 17). Thirty-eight of the 54 animals caught were whitefooted mice (Peromyscus leucopus). Twelve pine voles (Pitimys pinetorum) were caught in Area II, within an open canopy area of the woods which was

dominated by nimblewill (Muhlenbergia schreberi) grass and blackberry (Rubus allegheniensis) bushes. One pine vole was also caught in Area III. A total of four short-tailed shrews (Blarina brevicauda) were also caught on site.

Grid IA exhibited the highest trap success (9%), with the majority of captures consisting of P. leucopus. Grid I, within the same area but south of the railroad tracks, had the lowest trap success (0.02%) of all the areas.

## 3.3.3 Age, Sex, and Anatomical Metrics

Of the 38 Peromyscus trapped, 34 were adults, 3 were subadults and one was a juvenile animal. The mean body weight of adults was 22.3 g, as opposed to 15.94 g for the three subadults, and 11.80 g for the juvenile animal. Of the 34 adult Peromyscus, 19 were males and 15 were females. Two of the three subadults and the juvenile were females. Age and sex data for Pitimys and Blarina are included on the small mammal data sheets in Appendix J.

Review of body and organ metrics recorded reveals few apparent differences between trap areas (Table 18). Only total weight and total adrenal weights were normally distributed (Appendix G).

ANOVAs conducted on total weight and adrenal weight revealed no significant differences between areas (F=0.46, 2 d.f., p>0.64, and F=0.74, 2 d.f., p>0.48 respectively). ANCOVAS (see Section 2.3.4) revealed no significant differences between areas for either liver or kidney weight data (F=0.19, 2 d.f., p>0.82, and F= 1.84, 2 d.f., p>0.17 [Appendix G]).

The remaining organs (thymus, spleen, testes, and uterus) had weight data for which the error was not normally distributed. A non-parametric test (Kruskal-Wallis Test) revealed no significant difference between areas for any of these organs (Appendix G).

## 3.3.4 Pb Accumulation in Peromyscus leucopus

Individual *Peromyscus* trapped in the vicinity of the site contained Pb in their tissues at levels ranging from 0.2 to 13.0 mg/kg wet weight (Table 19, and Appendix E). Percent moisture ranged from 27 to 72% (Appendix E). An ANOVA comparing whole

body wet weight Pb concentrations of trapping areas Peromyscus between revealed significant differences in uptake (F=3.7, 2 d.f., p<0.10). This relationship held true when all animals, including the one juvenile and three subadults were considered (F=5.86, 2 d.f., p<0.01). Data in both data sets were first log transformed, since they were not normally distributed. mean comparison test showed the log transformed mean Pb concentration of Area I was significantly lower than the mean concentration in Area III. Mean whole body wet weight concentrations were 1.60, 3.10 and 4.77 mg/kg for Areas I, II and III, respectively. The mean for Area II was not significantly different from either Area I or III.

However when Pb concentrations were converted to dry weight as a means of standardizing the data to account for differences in moisture content, no significant differences between areas were noted. This was true when adults were considered alone (ANOVA, F=0.96, 2 d.f., p>0.39), as well as when all animals (adults, subadults, juveniles) were considered (F=2.45, 2 d.f., p>0.1).

Pb concentrations for the entire data set were compared with whole body wet weight. The correlation between log transformed whole body wet weight Pb concentration and total fresh body weight yielded an r value of 0.35.

## 3.4 Terrestrial and Wetland Habitat Assessment

Site vegetation may be categorized into four major vegetative communities: (1) palustrine broad-leaved deciduous forested wetland, (2) palustrine emergent wetland, (3) mixed deciduous forest, and (4) early successional old field (Figure 9).

The palustrine broad-leaved deciduous forested wetland is associated with the East and West Stream drainages, and was delineated as part of the RI<sup>(1)</sup>. This community is dominated by red maple and sweetgum trees, which appear to be about 20-60 years old. The understory of this community varies throughout the site. Along the West Stream north of the railroad tracks and along the southern third of the East Stream a dense thicket of greenbrier (Smilax rotundifolia), wild grape (Vitis sp.), and Southern arrowwood (Viburnum dentatum) is present. Along the West Stream south of the railroad tracks and throughout much of the East Stream area (Figure 9) a more open understory of the latter species is present. Sweet

pepperbush (Clethra alnifolia) is also dominant in more open, but saturated areas. In the herbaceous layer of areas prone to continuous saturation, such as the East and West Stream areas well south of the railroad tracks, skunk cabbage (Symplocarpus foetida) is common. In saturated areas with an open canopy, the skunk cabbage is replaced by arrow-leaved tearthumb (Polygonum sagittaria).

In addition to supporting populations of mice, voles, and shrews, the latter community provides habitat for larger species of mammals, such as red fox (Vulpes vulpes fulva), woodchuck (Marmota monax), striped skunk (Mephitis mephitis), raccoon (Procyon lotor), opposum (Didelphis virginiana), and white-tailed deer (Odocoileus virginiana). With the exception of the opposum, sign or tracks of all these species were observed during the field investigation. This woodland area can also be expected to support populations of breeding birds (Appendix K - List of Species Observed). Collectively small mammals and birds on site provide food for predators such as the northern black racer snake (Coluber constrictor), noted as common on site, and the red-tailed hawk (Buteo jamaicensis).

Surrounding this wetland at a slightly higher elevation is an upland deciduous woodland. This woodland is also dominated by sweetgum, but with other species more tolerant of drier conditions such as black cherry (Prunus virginiana) and white oak (Quercus alba) are present in the canopy as well. Within Area I, the wetland boundary is very abrupt in some locations and appears to be influenced by past excavative or earthmoving activities. In open areas and within canopy gaps of this forested herbaceous species indicative of disturbed conditions are not uncommon. An extensive (Figure 9) stand of Phragmites was noted along the west side of the former facility, while Virginia knotweed (Polygonatum virginicum) and pokeweed (Phytolacca americana) were not uncommon in the woodland east of the Exxon facility where small mammal Grid II was placed. Additional evidence of past human disturbance are numerous small mounds and gullies, apparently created from past earth-moving activities.

Palustrine emergent wetland (PEM) is limited largely to the area on both sides of the railroad tracks immediately west of the former facility, and to an area of the West Stream near Route 130 (Figure 9 and Appendix L - Site Photos). In mid-June, this entire area was inundated with less than one ft of water, but by the end of the summer it was nearly dry. North of the railroad tracks,

this community is dominated by spikerush (Eleocharis obtusa), with lesser amounts of tickseed sunflower (Bidens aristosa) and nimblewill grass.

South of the railroad tracks, the PEM community is dominated by nimblewill grass, and water smartweed (Polygonum sp.). Soft rush (Juncus effusus) is present in smaller amounts. This area appears somewhat degraded, as evidenced by a reddish-colored sediment and a discharge pipe, presumably from either the NL or the Exxon facility. Nevertheless, both killdeer (Charadrius vociferus) and common egrets (Casmerodius albus) were observed foraging in this area during the site investigation. The palustrine emergent wetland area near Route 130 is dominated almost exclusively by Phragmites sp.. A common egret was observed foraging in this area as well.

Both forested and emergent wetland areas on site also support turtles, as well as amphibians such as salamanders and frogs (Appendix K). These in turn are consumed by species such as the common egret and green-backed heron (Butorides virescens), the latter being observed on site as well.

An early successional old-field community occupies much of the area of the site in the immediate vicinity of the former facility, as well as the power line right-of-ways and edge of the rye field east of the Exxon facility (Figure 9 and Appendix L). On the western side of the facility, this community exists along the edge of the aforementioned sweetgum woods and consists primarily of little bluestem grass (Andropogon scoparius) intermixed with scattered forbs such as pussytoes (Anaphalis margariticea), Queen Anne's lace (Daucus carota), yarrow (Achillea millefolium), winged sumac (Rhus copallina) The power bushes, and scattered black cherry saplings. line areas along the eastern side of the Exxon facility support a thick growth of wild blackberry bushes with a variety of weedy forbs such as Joe-pye weed (Eupatorium purpureum), goldenrods (Solidago sp.), and grasses. This area is also disturbed, probably as a result of brush clearing for power line maintenance. A small uneven berm exists along the length of the East stream as evidence of past ditching activities.

#### 4.0 DISCUSSION

### 4.1 Aquatic Study

### 4.1.1 Pb Contamination in Sediment

Pb values in sediment as measured by XRF were consistent enough with analytical results from the RI<sup>(1)</sup> to confirm general trends of Pb contamination for selection of toxicity evaluation samples, and to characterize exposure levels for green frogs. In the West Stream area between the railroad tracks and Pedricktown Road, high Pb concentrations were recorded ranging from 4,000 mg/kg to above 20,000 mg/kg. These levels are similar to those previously reported. However, the high Pb concentrations in this study were recorded further north than in the former study, close to prior sampling location WS-10.

Results from sample locations 1, 3 and 5 agreed well with the RI. Location 1 was in the East Stream at the power line crossing, and corresponded approximately with location ES-5 in the RI. In the present study, XRF data varied from 386-576 mg/kg at this location, while it had been recorded at 206 mg/kg at ES-5 in the RI<sup>(1)</sup>.

Location 5, at the East Stream just south of the railroad tracks, roughly corresponded with prior location ES-7. In the present study Pb was recorded at 53 mg/kg at this location, while it had been recorded at 44 mg/kg at 3-6" bgs at ES-7 in the RI.

Location 3, located in the East Stream woods southeast of the powerline crossing (Figure 3) was situated about 200 ft north of ES-6 in the RI. In the present study, Pb concentrations were recorded at 32-37 mg/kg at this location, while in the RI they were recorded at 36.9 mg/kg at ES-6.

However, variability in results was observed at other locations. In the headwaters of the West Stream south of Pedricktown Road, higher Pb values were obtained in this study than in the RI. At location 4, which approximately corresponds with location WS-13 in the RI, Pb was recorded at 1100 mg/kg in the present study, versus 171 mg/kg recorded at the surface during the RI.

The variability in the results of the two investigations can be expected to arise from differences in sampling and analytical techniques, level of effort, and natural heterogeneity of Pb-contamination in the matrix. Stream conditions in

the interim period between studies may have also affected the fate of Pb in the sediment throughout the East and West Stream drainages. Despite this variability, it is clear that the most highly contaminated sediment was collected from the area located along the West Stream between Pedricktown Road and the railroad tracks. It is also clear that overall Pb contamination is greater in the West Stream than in the East Stream drainage.

### 4.1.2 Toxicity and Bioavailability of Pb in Sediment

Mortality of midge larvae (Chironomus tentans) exposed to site sediment was not directly related with sediment Pb concentrations. The low survival specific occurred within exposures at concentrations of Pb suggesting that other factors as well as potential Pb interactions with other variables were responsible for the observed mortality. Evaluation of changes the which occurred in the test water conditions during the test suggests that the mortality observed may be a result of loss of water alkalinity with subsequent pH depression, potential increased availability of Pb due to lower pH condition or other unmeasured factors. The high (86.7%) survival observed in the control group suggests it is unlikely that the observed results are due to an experimental artifact. Additionally, no significant mortality occurred in the exposures to the highest sediment Pb levels.

Chronic effects were found in the exposers where mortality did not occur. Reduced growth, as measured by changes in length and weight, was found between the control and the two highest sediment Pb exposures. It can then be stated that the under normal water quality conditions chronic effects on midges do occur at a level of about 1100 mg/kg.

The bioavailability of Pb and other heavy metals is influenced by sediment variables, such as TOC or grain size, since fine particles and organic material in the sediment may bind with the Pb and make it unavailable for uptake. In this study, no apparent relationship existed between toxicity and TOC or grain size. Survival was low at locations 2,5, and 9, the sediments from these locations had TOC ranging from 4.8 g/kg at location 5 to 160 g/kg at location 2. Furthermore, TOC values at locations 2 and 8 were similar, yet survival

differed by 80%. Similarly there was no clear association of test results to grain size distributions.

Water quality measurements from the dilution water suggest that alkalinity and pH may have had a profound effect upon the test results. alkalinity and pH values were notably higher, and not significantly altered from initial conditions, in the three groups exhibiting high survival (see Section 3.14, and Table 6). Inspection of physiochemical parameters measured from toxicity test water used in the toxicity evaluation (Table 6) reveals notable differences in both pH and alkalinity between locations where survival was low and those where it was high. At locations 2, 5 and 9, survival ranged from 0-15%. In each of these samples, the pH dropped below 5 during some point in the test. At the remaining locations, where survival exceeded 86%, the pH was above 5.2 units throughout the test.

Alkalinity values recorded during the test ranged from a value of <4 mg/L to a maximum of 12 mg/L in the samples exhibiting low survival (locations 2,5,9). In the samples showing high survival, alkalinity values equalled or exceeded 16 mg/L throughout the test and ranged as high as 56 mg/L.

C. tentans is relatively metals tolerant (6-7), and while species differences exist, Chironomids in general (8-10) are believed to be metals tolerant. The selection of this species as the test organism was based upon the system being evaluated, one in which Chironomids would be anticipated to be a major component.

It is generally accepted that pH changes effect the uptake of metals from macroinvertebrates (11,12). Un sediments Under equilibrium conditions with a temperature equal to 25°C and pressure at 1 atmosphere (atm), the solubility of Pb increases steeply below a pH of 8 units. Between a pH of 5 units and a pH of 4 units, the solubility of Pb is predicted to increase nearly 10,000 times (13). Thus, the depression of the pH within the test exposures may have also contributed the test results, due to increased availability.

Evidence of Pb release is found in the data collected in the RI<sup>(1)</sup> on Pb concentrations in

sediment and surface water at the same location points. The Pb concentration of the surface water was negatively correlated with the pH of the sample, suggesting that as the pH is lowered the Pb may be released from the sediments. This is apparently true over a range of sediment Pb concentrations (Appendix C) ranging from <5 mg/kg to more than 4,000 mg/kg.

### 4.1.3 Toxicity Test Conditions Versus Field Conditions

Water quality parameters measured by the Hydrolab Surveyor II in the field at three locations (East Stream Pedricktown Road, West Stream Pedricktown Road, and West Stream Route 130) were within the same range of values reported by the toxicity testing laboratory for the toxicity testing water. For example, the Hydrolab reading for pH was 5.91 at the West Stream, Pedricktown Road. This corresponded approximately with location 4, where toxicity testing water had a pH range of 5.5 to 6.7 units during the laboratory study.

Sediment pH values measured in the field using an electronic pH meter were generally higher than values recorded in toxicity testing water. could be caused by stirring the sediment during Also, laboratory procedures. the laboratory a relatively static represents system controls for much of the environmental fluctuations which would normally occur in nature. results may have implications for site remediation activities, since disturbing the conditions may alter the environmental conditions of the sediment and thereby alter the release or bioavailability of the sediment bound Pb.

Dissolved oxygen values measured in the field were generally lower than laboratory measured values, although measurements were not taken at the same exact locations. It is likely that some of this variation is because the laboratory aerated the toxicity testing chambers, which was required by the test protocol. However, fluctuations in DO values between dates and locations are also evident in the Hydrolab data and are interpreted largely as a result of drawdown in the streams. During the aquatic study in August, the East Stream was significantly drier than during the preliminary

### 4.2 Pb Accumulation in Green Frogs

#### Comparison with Literature Values 4.2.1

the present study the mean wet weight concentration of green frogs was 8.2 mg/kg. maximum concentration was 23 mg/kg, in a frog from the West Stream; the minimum concentration was 1.4 mg/kg in a frog from the West Stream below Rt. 130.

Few data are available from the literature on concentrations of Pb in anurans. Pb concentrations ranging from 6.4 to 14 mg/kg dry weight were reported (14) for four wood frogs (Rana sylvatica) collected 9.7 km from a zinc smelter plant. If the lowest percent moisture value (74%) recorded for frogs in this study is used, then the wet weight concentration of the most contaminated frogs in the above study (14) would be about 3.6 mg/kg wet weight.

Wet weight concentrations of up to 300 mg/kg Pb were reported in bullfrogs (R. catesbiana) collected downstream of a mining site in Missouri (15). Upstream frogs in the same study had a background mean concentration of 1 mg/kg and a maximum concentration of 6 mg/kg. In comparison, seven out of the 13 frogs collected in the present study had wet weight Pb concentrations less than 6 mg/kg.

Background organ levels were reported (16) for leopard frogs in a rural area in Vermont. Wet weight liver concentrations ranged from 1 to 6.2 mg/kg, while kidney concentrations ranged from 1.3 to 8.2 mg/kg. Whole body concentrations would be expected to be lower, since Pb in anurans is primarily stored in the liver, kidney, and bone (17).

Other studies have focused on the uptake and effects of sediment and surface water contamination on tadpoles. Green frog tadpoles collected in an area with a mean concentration of 66 mg/kg Pb in sediment (18) had a mean wet weight tissue concentration of 14 mg/kg. Sediment Pb concentrations and tissue Pb concentrations were positively correlated.

A mean dry weight Pb concentration of 4139 mg/kg was recorded in Rana tadpoles collected from a Missouri tailings pond<sup>(19)</sup>. Tadpoles collected 1 km and 25 km downstream had mean dry weight Pb concentrations of 552 and 37 mg/kg respectively.

In a study of the effects of Pb on green frog tadpoles<sup>(20)</sup> no significant difference was noted in avoidance of Pb or in locomotor activity under increasing Pb exposures. However, the authors reported increased variability in locomotor activity in tadpoles exposed to 750 ug/L and 1000 ug/L Pb versus 500 ug/L and control groups as evidence of the effects of Pb.

Strickler-Shaw and Taylor<sup>(21)</sup> found that sublethal exposure to 750 ug/L Pb adversely affected acquisition learning and memory in green frog tadpoles. These concentration levels were exceeded by some of the values reported in the RI for surface water in the East and West Streams(1).

Other researchers<sup>(22)</sup> have found that an injection of 1 ppm Pb<sup>2+</sup> into Bufo arenarum eggs resulted in embryonic malformations such as failed closure of the neural tube, small and cylindered tails, reduced body size, and incurvations in the body axis.

### 4.2.2 Tissue and Sediment Pb Concentrations

Frog tissue levels generally reflected sediment Pb concentration ranges in the areas where they were captured. However, only qualitative comparisons were made as these sample sizes were too small for statistical comparison. Green frogs collected in the West Stream drainage generally followed the gradient of Pb contamination in the sediment (Table Whole body wet weight tissue concentrations for the two frogs caught at Pedricktown Road were 2.3 and 3.9 mg/kg, where sediment was measured at approximately 1000 mg/kg. Further north along the drainage, directly across from the NL facility, whole body tissue concentrations were higher, ranging from 5.2 to 23 mg/kg Pb. Sediment concentrations in this area were measured at greater than 4000 mg/kg Pb. The two frogs caught at the intersection of the West Stream and Route 130 had whole body tissue concentrations of 1.4 and 8.6 mg/kg, respectively. Sediment concentrations in this area were again close to 1000 mg/kg Pb.

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tissue concentrations body frogs the inhabiting East Stream drainage near Pedricktown Road appeared comparable to those of the West Stream near Pedricktown Road with the exception of one individual which had a Pb 13 mg/kg (Table 7). concentration of individual was the largest frog of any caught in the study and could have been the oldest and thereby subjected to a longer exposure period or could have moved from a more heavily contaminated area.

Frogs inhabiting the site can be expected to accumulate Pb through contact with Pb contaminated water and sediment, as well as through their diet. While specific relationships between these media and frog tissue levels can not be made; comparisons can be made with sediment Pb concentrations as indicative of overall exposure potential.

Since all sediment samples were taken within 50 feet of the frog capture points, the sediment Pb data should give a reasonable idea of the Pb concentration to which the individuals were exposed in their home ranges. Green frogs exhibit homing behavior when displaced from their home ranges at distances up to 600 meters (m) (23). Also, the home range is usually quite limited (23-24). Martof (24) found green frog home ranges to extend from 20 to 200 square meters (m²), with a mean of 61 m². However, approximately 85% of the frogs in his study were recaptured within 10 m of their original location.

Oldham<sup>(23)</sup> conducted a recapture study and noted that green frogs moved a mean distance of only 37 m from their original point of capture to their recapture point. Ryan<sup>(25)</sup> observed that many green frogs were recaptured within a few feet of their original capture point, and with few exceptions they remained in the same pool, pond, or backwater from one season to the next.

### 4.2.3 Age Structure and Pb Uptake

Comparison of weight and length data on frogs collected in this study (Table 7) with literature values (25-28) indicates that most if not all individuals collected were less than two years old, and several may have undergone transformation from tadpoles shortly before capture. Martof (26) noted a mean length at transformation of 32.6 mm, with

values ranging from 28.4 to 36.4 mm. Ryan<sup>(25)</sup> recorded a mean length at transformation of 32 mm, with a range of values from 26 to 38 mm. Pough and Kamel<sup>(27)</sup> noted that green frogs in their study weighed 3 g at transformation. Based upon these criteria, at least six of the 13 frogs in the present study had probably metamorphosed recently, as evidenced by body weights close to 3 g and total length measurements of 35 mm or less.

Green frogs spend a maximum of 8-10 months in their larval stage before transformation<sup>(24)</sup>. However, they are capable of reaching maturity within 90 days, and will often mature within a single season<sup>(26,27)</sup>. Martof<sup>(26)</sup> noted that individuals grew by a mean of 36.4 mm one year later, which implies the average frog at one year and ten months would measure about 69 mm in length. The frogs collected in this study all measured less than 60 mm.

The studies reviewed (24,26) noted that when green frogs generally reached about 60 mm in length, they left their subadult habitat (characterized by shallow water with marsh vegetation) and began to use deep pools within streams. Results of the present study support this finding. The East and West stream drainages offer little deep pool habitat over most of their extent, and the frogs caught were small subadults. Larger frogs observed or captured in this study were confined to three deep pools located within the West Stream near Pedricktown Road and at Route 130, and within the East Stream near Pedricktown Road.

### 4.2.4 Food Chain Effects

Green frogs are preyed upon by a variety of predators that inhabit or potentially inhabit the site, including wading birds, snakes, snapping turtles (Chelydra serpentina), raptors, raccoons, skunks, opossum, mink (Mustela vison) and red fox<sup>(29)</sup>. Raccoon tracks were noted and both the green-backed heron and common egret were observed on site. The northern black racer was commonly observed on site as well, and a snapping turtle was observed in the immediate vicinity of the site. Fox sign was also noted along the railroad tracks.

Green-backed herons collected downstream from a Missouri mining site<sup>(15)</sup> were reported to have had a mean wet weight Pb level in their livers of 0.5

mg/kg, while bullfrogs collected from the same stream accumulated close to 13 mg/kg fresh weight Pb, however, it is not possible to directly evaluate the relationship between the herons and the frogs in that study.

Northern water snakes (Nirodia sipedon) accumulated less than 10 mg/kg whole body Pb in the same study<sup>(15)</sup>. Few other data are available for other potential frog consumers, although Pb does not normally magnify up the food chain<sup>(15,30)</sup>.

At the biochemical level, Pb has been documented to depress delta aminolevulinic acid dehydratase (ALAD) activity, an enzyme essential for hemoglobin synthesis<sup>(30)</sup>. While a multitude of studies have shown decreased ALAD levels in a variety of species, the significance of this decrease is debatable<sup>(30)</sup>.

At high concentration levels, Pb is detrimental to nervous system function and may cause liver and kidney problems, while chronic exposure can cause learning deficiencies and otherwise influence behavior (30).

### 4.3 Earthworm In-Situ Bioaccumulation Study

### 4.3.1 Earthworm Survival

Eisenia survival was notably low at locations 11, 12, and 18-20 (Table 13). This could be due to several factors either interacting or working independently, including Pb concentration, handling of the worms, individual differences between worms, grain size, percent organic matter, percent moisture, or predation. Additionally, the reasons for mortality may have differed between locations.

It is interesting to note that at some locations (2, 3, 6, 11) where mortality was observed, worms surfaced and desiccated at the surface of the chamber soil. This may suggest direct avoidance of the soil. At other locations such as location 20, holes on the surface of the soil were observed, indicating that the worms had been surfacing regularly. Yet, at the end of 28 days, no earthworms were found in the chamber. Based on test chamber design, and observations of earthworms during trial chamber tests, it is highly unlikely that any worms escaped from the chambers.

Also, it is unlikely that temperature significantly affected the worms, since soil temperature should have been reasonably uniform between locations, varying slightly with percent moisture in the soil. With the exception of chamber location 6 all chambers were installed under a full canopy, and none were exposed to direct sunlight (Chamber 6 exhibited 70% survival). Grain size less than optimal at nearly undoubtedly locations in the study, but is not correlated with test mortality. Soils throughout the study area consisted primarily of sand. Percent moisture values were seldom lower than 10% in the chambers, and greater than 30% in 9 out of 20 of the There was no correlation between chambers. mortality and percent moisture, and the worms in chamber 20 which had been exposed to a moist silt loam soil had 0% survival. Prior studies have that earthworms will tolerate limited shown desiccation, experiencing greater than 50% weight loss and later recuperating (31).

Precipitation followed by brief flooding may have occurred in some chambers, but it is interesting that worms at location 17 were immersed in water when retrieved, and appeared healthy. Earthworms will often surface when soil conditions become saturated, yet they can survive immersion in water for up to 30 days<sup>(31)</sup>.

The mean weight of worms in all chambers decreased significantly during the exposure period (see Section 3.2.4), which suggests that the worms may have been starving, possibly as a result of avoidance, or low food availability or quality. Starvation may have been directly responsible for mortality in some cases. Excess organic material can be detrimental in some cases, since Eisenia exposed to soil which is too rich in protein may exhibit a toxic syndrome known as sour crop<sup>(32)</sup>, however, this is unlikely to have occurred during this study.

Predation may have also caused mortality in some chambers. An immature red-backed salamander (Plethodon cinereus) was found in chamber 20 at the end of the study, and no worms were found. This could be an example of predation, but it is unlikely that mortality observed in the remaining chambers can be ascribed to predation.

Pb contamination acting independently can be discounted, since several other chambers had higher Pb concentrations than these (Table 13). However, the observed mortality may have been a consequence of several interacting factors which could influence the availability of Pb for uptake. In one study<sup>(33)</sup> the LC50 value for *Eisenia* exposed to artificial soil was greater than 5,000 mg/kg soil Pb.

### 4.3.2 Tissue Pb Concentrations

In this study, Pb was accumulated by earthworms at concentrations well below those in the soil. This is consistent with results from other investigations  $^{(34-38)}$ , which used a variety of worm species. Only two studies were found which noted that earthworms are capable of storing Pb in higher concentrations than the soil they inhabit  $^{(39,40)}$ . In both of these studies soil concentrations were low. In the first study  $^{(39)}$ , soil Pb concentrations were  $^{(2)}$  mg/kg, while in the second one  $^{(40)}$  they were 170 mg/kg or less.

Worms in the present study accumulated a maximum of 170 mg/kg despite the fact that they were exposed to very high concentrations of Pb in the soil. Review of bioconcentration factors revealed that the proportion of Pb accumulated by worms decreased exponentially as soil Pb concentration increased (Table 13, and Appendix I). This could be a result of avoidance of Pb ingestion at higher soil Pb levels or a function of increased Pb excretion at higher tissue levels. Alternatively, the Pb at higher concentrations may have been less available (see Section 4.3.4.2 below).

A review of the literature reveals a similar trend in several studies. Ma<sup>(41)</sup> reported that BAF values for metals generally declined with increasing soil concentration. In his study, *L. rubellus* exposed to 20 mg/kg Pb accumulated the same amount of Pb as individuals exposed to 3000 mg/kg. Worms exposed for a 12-week period to 3000 mg/kg Pb actually accumulated less Pb than those exposed to 20 mg/kg.

Other researchers have reported relatively high BAF values. In one study  $^{(34)}$ , values ranged from 0.66 to 0.96 for worms (species not noted) exposed to soils with low (16-43 mg/kg Pb) concentrations. In another study  $^{(36)}$  BAF values were recorded for a variety of worm species (but not *E. foetida*)

exposed to 147 mg/kg Pb. BAF values were 0.30 for Lumbricus terrestris and ranged as high as 0.57 for Allolobophora chlorotica. The values recorded for Eisenia in this study (0.16-0.25) at a similar concentration range (180-190 mg/kg soil Pb), were slightly lower, although species variability, soil characteristics, and individual differences do not make the results directly comparable.

Van Hook<sup>(42)</sup> reported BAF values ranging from 0.11 to 0.30 for several earthworm genera collected from soils ranging in Pb concentration from 18 to 50 mg/kg. Another study(40) reported BAF values of 0.36, 0.47, and 0.55 in worms exposed to silt loam soils with 130-170 mg/kg, 30-64 mg/kg, and 20 mg/kg Andersen<sup>(38)</sup> reported that "on Pb, respectively. the average" L. terrestris worms in his study concentrated Pb by a factor of 0.24. Most of the worms in his study were exposed to <23 mg/kg soil Pb, but some were exposed to nearly 2000 mg/kg. This figure is close to the 0.23 BAF value of Eisenia in the present study, which were exposed to <500 mg/kg Pb.

Although BAF values were in some cases higher in other studies, the maximum concentration of Pb found in earthworm tissue in this study (170 mg/kg wet weight) is higher than in most (34,35,37,40-45). However, these studies used different worm species, and were exposed to far lower soil concentrations.

Comparison of Pb concentrations of Eisenia to a prior study using the same species showed that levels accumulated in this study were higher than reported in some other studies. Hartenstein, et al., (44) exposed Eisenia foetida to activated sludge with 180 mg/kg Pb for a period of four weeks and reported a mean dry weight tissue concentration of 99 mg/kg, with a percent moisture content averaging 81.8%. This would yield a wet weight concentration of 18.02 mg/kg Pb. In contrast, earthworms in the present study exposed to 180 mg/kg for four weeks accumulated 29 mg/kg Pb wet weight, and those exposed to 190 mg/kg accumulated 48 mg/kg.

Eisenia foetida exposed to Pb contaminated sediment (135 mg/kg) for seven days in another study (43) had a mean tissue concentration of 5.13 mg/kg dry weight. The mean concentration of that group was

actually less than their "background" Pb concentration in worm tissue of 5.23 mg/kg dry weight.

### 4.3.3 Time Dependent Accumulation

The stock Eisenia used in this study did not contain Pb above a detection limit of 0.25 mg/kg. At location 4, worms were analyzed after 10, 20, and 28 days, and were found to contain 15, 21, and 29 mg/kg, respectively. Thus, uptake was rapid during the first ten days, and was still increasing at the end of the 28-day exposure period. indicates that the accumulation had not yet reached steady state, however, based on the accumulation studies cited above and the shape of accumulation curve, it is doubtful that tissue concentrations would be dramatically higher than that observed at 28-days. The observed results do suggest that the tissue concentrations reported in this study should be viewed as minimum estimates, of steady state concentrations for the exposures utilized.

(44) Hartenstein, et al., reported tissue concentrations of Eisenia exposed to 180 mg/kg Pb at the initial time of the study and after 14 and respectively. The initial days, concentration of the worms they used was very high (63 mg/kg dry weight), and the concentration measured after 14 days was actually lower (56 After 28 days, the worms had accumulated mg/kg). mg/kg Pb. Because of the high initial concentration of Pb in their stock worms, it is difficult to evaluate their results.

### 4.3.4 Soil Characteristics and Bioaccumulation of Pb

### 4.3.4.1 Surface Pb Concentrations

Actual field exposure to earthworms is complex as surface soil concentrations on site vary greatly over any given area and with depth (Table 9). Lead concentrations from 0 to 1" bgs were substantially higher than concentrations farther below. This distribution pattern occurs in both upland and wetland locations.

Airborne transport of Pb could offer an explanation for the "spotty" nature of the Pb

concentrations in some areas. Airborne particulates could easily lodge on the leaf surfaces of surrounding trees and then become incorporated into the organic layer after the leaves fall to the ground. Also, storms, high wind events or other atmospheric conditions could have interacted with facility operations resulting in the transport of contaminated particles or slag material from the piles still present on site.

Grain size results show the majority of onsite soils to consist of sand. These results are consistent with the description of the Sassafras-Galestown-Woodstown loamy complex mapped on site by the U.S. Soil Conservation Service (46). This soil has been described as highly subject to wind erosion<sup>(46)</sup>. This may be an additional cause of the variability in surface soils.

Another potential influence on the variability of surface Pb concentrations is prior soil moving activities. Small, abrupt mounds were noted in several areas of the site, with gray hardened soil indicative of lower surface horizons. Ditches and berms were noted throughout the site. This prior disturbance may have moved surface soil, where Pb contamination was concentrated, into smaller, more concentrated areas.

The XRF data collected in this study also suggest that Pb is largely bound to the humic layer, which has been suggested by previous studies (47).

# 4.3.4.2 Soil Characteristics Influencing Bioavailability of Pb

Tissue concentrations of Pb were not correlated with soil Pb concentrations, which suggests additional variables affected Pb uptake in the chambers. The bioavailability of Pb is likely influenced by a number of factors in the soil, including pH, grain size, organic content, and percent moisture of the soil. The step-wise regression analysis did not demonstrate that these factors were good predictors of the Pb content in earthworm

tissue. Only percent organic matter was significantly (at the p<0.1 level; p=0.06) associated with the Pb tissue level.

Several explanations can be offered for this lack of correlation of Pb uptake with soil characteristics. One is that sample sizes may have been insufficient to detect any trends. Secondly, earthworms may have shown individual variation in their responses contamination: this explanation is not mutually exclusive from the first. Finally, the observed differences in uptake could be due to additional variables such as cation exchange capacity (CEC), or unknown variables which were not measured.

Prior studies have assessed the contribution of the above variables influencing Pb uptake. et al., (48) conducted a step-wise regression using Pb uptake as the dependent variable and found that both low pH and percent organic matter significantly increased Pb uptake in Lumbricus rubellus. Morgan and Morgan<sup>(35)</sup> conducted a similar regression and found that pH, cation exchange capacity, and soil calcium (Ca) had a major influence on Pb accumulation in L. rubellus. The pH levels ranged from 3.5 to 6.1 in the first study, and from 4.3 to 7.8 in the latter study. In the present study, pH values were consistently close to neutral, ranging from 6.2 to 7.0 (Table 10).

Several other studies have emphasized the importance of pH in affecting Pb bioavailability. Kiewiet and  $Ma^{(49)}$  reported that acidification decreases soil adsorption of heavy metals, resulting in a greater concentration in the soil solution, and hence greater availability to earthworms.  $Ma^{(41)}$  found that pH and cation exchange capacity were both significantly negatively correlated with Pb uptake in L. rubellus.

In contrast, Andersen<sup>(38)</sup> found that pH had little effect on Pb uptake in Allolobophora and Lumbricus earthworms, although pH values in his study ranged from 5.7-6.0, which were closer to those recorded in the present study.

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Thus, that Pb availability is complex and many soil factors can potentially control the availability in any given soil.

Beyer<sup>(34)</sup> reported that applying lime to soils with an initial pH ranging from 4.6 to 5.9 had little influence on uptake in earthworms. The species of worms used in their study were not identified.

Grain size and the organic matter content of soil are known to influence uptake of Pb because smaller particles tend to bind the Pb ions more tightly. In one study (41), Pb was better accumulated by L. rubellus in sandy versus loamy soils. Andersen (38) stated that heavy metals are strongly immobilized in soil by adsorption to clay minerals and interaction with organic matter, resulting in low concentrations in soil water.

While some variability is evident (Table 11), the grain size in soil in all chambers consisted primarily of sand. The silt/clay fraction of the soil, critical for binding Pb, was between 5 and 12% in all but four of the chambers. The silt/clay percentage in the chambers was not correlated with Pb uptake in the worms.

Percent organic matter (Table 12), however, varied greatly between chambers. Organic matter was removed by ignition prior to grain size analyses, such that in some cases the analyses were based upon less than 25% by weight of the sample submitted for analysis. Percent organic matter is probably a more realistic indicator of the organic material in the chamber which could be expected to bind Pb than is the total organic content (TOC), although the two parameters were highly correlated.

The percentage of organic material was significantly less for samples collected on the eastern side of the site (locations 1-7) which reflects the fact that several of the chambers on the western side were located within or along the wetland boundary. Percent organic matter ranged from 4-22% in chambers 1-7, while organic matter in chambers 8-19 ranged from 30-78%. Also, the appearance of

the forests in the two areas was different, as the eastern side had a more open canopy and understory than the western side. This may explain some of the observed difference in organic matter, since surface litter would be less in the more open area.

This difference was observed in the field during installation of the chambers. High amounts of organic material were noted in several chambers on the western side, while on the eastern side of the study area, soils were noticeably sandier and drier. Percent moisture data collected during installation show that the eastern soils were generally drier than the western soils; in all cases they had <20% moisture (Table 10).

Despite the fact that soil Pb concentrations were substantially higher in the western chambers (mean = 3,127 mg/kg) than in the east side chambers (mean = 534 mg/kg Pb), the worms western side did not accumulate significantly more Pb than those on the eastern side. In fact, the maximum wet weight concentration of Pb noted in west side worms was 170 mg/kg, as opposed to a maximum of 130 mg/kg Pb wet weight in the east side chambers. These results collectively suggest that the high percentage of organic matter on the west side is acting to limit the availability of Pb for uptake. Morgan and Morgan (35) concluded that the relatively low concentrations of Pb found in L. rubellus in their study probably reflected the immobilization of Pb through binding to various soil inorganic and organic compounds.

The immobility of Pb relative to other metals has been noticed in several prior earthworm studies  $^{(33,34,36,41,49,50)}$ . A study using artificial soil and a two-week exposure period found that absorption of metals by earthworms from the soil was highest for Cadmium (Cd), while Cd was > Copper (Cu) > Zinc (Zn) > Nickel (Ni) > Pb $^{(33)}$ . Another study showed identical results, but did not investigate the Ni $^{(34)}$ . Ma $^{(41)}$  reported similar results except that Pb was higher in the worm tissue than Nickel (Ni), and he did not include Zn in his study. Kiewiet and Ma $^{(49)}$  reported that worms showed a higher uptake of Cd than Pb. Other

investigators found that the uptake of Pb in worm tissue differed from that of Zn and Cd in that it did not exceed the concentration in the soil (36).

### 4.3.5 Environmental Factors Influencing Pb Accumulation

According to Ireland(51), in temperate climates the maximum activity of earthworms in the soil is when soil temperature is between 4 and 11° C and when percent moisture is high. He further suggested that activity would have a positive influence on Soil temperatures measured at location 4 uptake. in the present study ranged from 16.8 to 25.9° C, and soil moisture was variable. While this may have negatively influenced uptake, it is unlikely that soil temperatures were detrimental to the This is assuming that soil temperatures recorded at location 4 were indicative of those in the remaining chambers. *E. foetida* placed in a water bath for 12 hours<sup>(52)</sup> exhibited a heat death temperature of 33° C, well above the maximum soil temperature in this study. Furthermore, Aston<sup>(53)</sup> stated that 25° C was the ideal temperature for cocoon production in Eisenia.

### 4.3.6 Toxic Effects of Pb Accumulation in Earthworms

### 4.3.6.1 Acute Toxicity

In a study comparing the toxicity of several chemicals to E. foetida, lead nitrate was classified as very toxic<sup>(32)</sup>. This classification was based upon an  $LC_{50}$  value ranging from 10-10,000 ug/cm<sup>2</sup> for Pb administered to worms for 48 hours on moist filter paper.

A study which tested two Pb salts (acetate and nitrate) using contact tests concluded that the two salts did not show a significant difference in toxicity to E. foetida<sup>(33)</sup>. The Pb acetate exposure generated a  $LC_{50}$  of 50 while the Pb nitrate exposure generated a  $LC_{50}$  of 64.

In contrast to contact tests on filter paper, earthworms seem remarkably tolerant of Pb levels in soil. In the above study  $^{(33)}$  E. foetida worms had an LC<sub>50</sub> value of 5941 mg/kg.

This value was exceeded at three of the earthworm chambers tested in the present study.

High tissue Pb concentrations have been reported elsewhere in the literature with no apparent effects to the individual worms. Ma, et al., (48) reported values up to 670 mg/kg in Lumbricus terrestris. These worms were depurated prior to analysis. Other studies have reported Pb levels as high as 2600 mg/kg in Lumbricid earthworms (see Ireland (51) for review), although it is not clear whether or not the worms were depurated.

### 4.3.6.2 Chronic Effects

Weight loss in test worms occurred in all chambers and was correlated with Pb concentration. Weight loss was greater than 50% (wet whole body weight) in some cases. A prior study (43) reported weight loss in E. foetida exposed to contaminated sediment to range from 16 to 56% versus 12% in their control group. In contrast, a laboratory study (54) found Pb to have little effect on the growth of E. foetida over a 20-week period.

The correlation between weight loss and soil Pb levels in the present study suggests that the worms may have avoided eating the Pb contaminated soil, or that Pb uptake may have interfered with nutrient absorption. However, the correlation value of 0.6 suggests that there is variability in the data set and that factors other than Pb may have influenced weight loss as well.

In several of the chambers located at the edge of the wetland on the western side of the facility (chambers 8,9,14,17-19) the organic material present in those soils may have presented an anaerobic food source unsuitable for worms, as was suggested in a study with similar wetland conditions<sup>(50)</sup>. While this would account for the weight loss in these chambers, this does not account for the weight loss observed in the remaining 14 chambers in the study.

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Despite the fact that *E. foetida* is known to feed on organic material, it is unlikely that weight loss was influenced by a lack of organic matter in the soil. Soil samples consistently revealed percent organic matter in excess of 5%, and upwards of nearly 80%. Marquenie and Simmers (50) found there was no correlation between organic matter content of the soil and weight increase in their study of *Lumbricus rubellus*. Moreover, the percent organic matter in several of the chambers was close to test conditions used in other studies (33,48).

Other potential chronic effects noted in this study were lesions and other morphological abnormalities observed in the worms upon removal from their chambers. However, these observations can not be linked to any parameters measured or other observations made.

Lesions in earthworms exposed to Pb have apparently not been described in literature. In fact, a study of metals effects<sup>(54)</sup> noted that Pb was the least toxic of the metals they tested (Cd, Cu, Ni, and Zn) with regard to effects on growth and reproduction in E. foetida. Yet, constrictions noted in this study are similar to what were reported in a prior study of E. exposed to N-methyl carbamate foetida insecticides (32). The authors noted swellings between segments 30 and 40, which often burst leaving a bloody ulcer that extruded fluid and eventually severed some worms. It is possible that these lesions may have been caused by the presence of another compound in the soil.

#### 4.3.7 Food Chain Effects

Earthworms are consumed by a variety of species of birds, mammals, small amphibians and reptiles<sup>(29,51,55)</sup>, and along with other invertebrates form an integral part of terrestrial food web. When the earthworm chambers were installed, Lumbricid worms (species unknown) were noted in the surface organic layer at some locations on site. Thus, earthworms component of the food web at this site.

Pb is not thought to magnify in terrestrial food chains (30), but consumption of Pb contaminated prey can result in Pb accumulation at higher levels of the food chain which could cause detrimental effects (17,30,33,51,56). Ma (56) reported that availability of Pb to earthworms was reflected in the liver and kidney tissue of moles (Talpa europea), one of their major consumers.

laevis) fed live earthworms (Xenopus containing 10, 308 and 816 mg/kg Pb accumulated variable amounts of Pb depending upon their feeding intake rate and exposure period (17). ingesting worms with 10 mg/kg Pb at a rate of 198 mg/day (wet weight) for eight weeks accumulated Pb from 0.59 to 5.00 mg/kg wet weight, depending on the tissue analyzed. The highest levels of Pb were accumulated in the bones, liver and kidneys. It is important to note that of the 10 mg/kg Pb present in the worms fed to the toads, only 5.63 mg/kg was present in the tissue. The remainder was in the soil contained in their alimentary tract (17). Thus. the levels of Pb in the worms they were fed were significantly lower than the tissue concentrations in worms in the present study.

### 4.4 Pb Accumulation and Effects on Small Mammals

### 4.4.1 Pb Accumulation in Peromyscus leucopus

### 4.4.1.1 Trap Success and Pb Accumulation

Peromyscus leucopus was chosen as the target species for Pb analysis based upon its relative abundance on site and its position in the food chain as an omnivore. Although three species of small mammals were trapped on site (P. leucopus, Pitimys pinetorum and Blarina brevicauda) trap success was low throughout, averaging 5% or less on all grids with the exception of Grid IA. Percentage trap success was somewhat inflated at Grid IA, since traps were left out for only one night and trap success is often greatest during the first trap night.

Nevertheless, the area within Grid IA represented excellent small mammal habitat, and trap success there contrasted greatly with that of Grid I, where only one animal was caught during the first trap night. The herbaceous vegetation in Grid I had undergone

considerable trampling during installation of earthworm chambers, and while vegetation had approximately one month to recuperate, this may have still affected trap success. doubtful that Pb contamination could account for differences in trap success between areas in this study, since trap success was not much different in Grid III (the most contaminated than the other grids. differences in trap success between areas are more likely а function of characteristics. Tissue and Soil Pb Concentrations

### 4.4.1.2

Significant differences were evident in wet levels of adult P. leucopus weight Pb collected in different areas of the site, but dry weight concentrations. differences, in wet weight means, generally differences surface reflected in concentrations between areas. Area I had a significantly lower wet weight concentration Area III. Area II had а concentration between the other two areas, but was not significantly different from either However, when data were normalized to dry weight in order to account for individual differences in percent moisture no significant differences between areas were noted. suggests that the analysis was very sensitive to relative small differences in the data set.

Whole body tissue values ranged from 0.2 to 13 mg/kg Pb wet weight over the entire site. The mean whole body Pb concentration in mice trapped in Area I was only 1.6 mg/kg, whereas that of Area III was close to 5 mg/kg. variability within Individual areas was evident. For instance, one individual each in Areas II and III had concentrations as high as 13 mg/kg.

Some variability could be a result from emigration from adjacent areas, trapping was conducted such that it would minimize the Additionally, induction of emigration. variability in tissue Pb content within areas may reflect (1) individual differences in uptake as a result of home range, and (2) the variability observed in surface concentrations throughout the site.

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The lack of significant differences in dry weight means between areas suggests that the above factors introduced sufficient variability to obscure differences between areas.

The above explanations are not mutually exclusive. The home range size of *P. leucopus* is variable, averaging approximately 0.1 hectares (57,58). Given the "spotty" nature of Pb contamination throughout the site, it is conceivable that individuals inhabiting the same grid area could be subjected to differing concentrations of Pb. Furthermore, surface soil concentrations appeared to differ between the three trap grid areas.

However, statistical comparisons between areas could not be made since XRF data collected along transects cover to representative area and not at random. Thus, while mean surface soil Pb concentrations variability differed between areas, the between areas may have superseded any apparent differences. Also, the characterization of surface Pb concentrations within each area was limited to less than 20 XRF points per area.

## 4.4.1.3 Comparison of Tissue Values With Prior Studies

Whole body tissue Pb concentrations in this study are comparable to or slightly higher than those reported in other studies of Peromyscus inhabiting heavily travelled roadsides or at a significant distance from smelters. Two of the mice in the present study had whole body concentrations greater than 13 mg/kg, but the majority of animals had concentrations below 7 mg/kg, and the mean site concentration was 3.16 mg/kg.

One study<sup>(14)</sup> reported whole body Pb concentrations in P. leucopus exposed to Pb contaminated soils located 2 and 10 km from a zinc smelter. They found that the mice exposed to 2700 and 490 mg/kg Pb had dry weight tissue concentrations of 17 and 7.4 mg/kg, respectively.

A second study of smelter contamination (59) reported data on deer mice (P. maniculatus).

Mice had whole body wet weight concentrations ranging from 23.5 to 173 mg/kg in one smelter area, and from 52.7 to 58 mg/kg in another. The authors suggested that these levels were sufficient to incur adverse effects in the mice.

Getz, et al., (60) found whole body Pb levels of 6.3 mg/kg dry weight in P. maniculatus collected within 10 m of highways. Their study concluded that concentrations in the mice were well below toxic levels.

In other studies of roadside Pb contamination, individual P. maniculatus had whole body tissue concentrations (dry weight) of up to 6.8 mg/kg in areas within 10 m from the road (57). Soil concentrations on the median strip of the highway were about 180 mg/kg. Another study of highway effects (61) found P. leucopus accumulated a mean of 4.91 mg/kg Pb from highway roadside habitats. A study of P. maniculatus in Colorado (62) found that mice exposed to a mean roadside Pb concentration of 73 mg/kg had Pb concentrations of 52.1 mg/kg dry weight in the bone, 8.46 mg/kg in the kidney, and 3.29 mg/kg in the liver. Whole body concentrations were not analyzed.

P. leucopus exposed to Pb from forest sites irrigated with a 299 ug/kg effluent sludge mixture had mean wet weight kidney and liver concentrations of 2 and 7.11 mg/kg, respectively (63). These values were higher than for mice collected at non-irrigated sites, but they concluded that these levels were not toxic.

### 4.4.1.4 Potential Effects of Pb

According to Eisler (30) the literature on Pb effects is abundant for domestic livestock and small laboratory animals, but noticeably lacking for feral mammals. A major difficulty in relating laboratory to field results is that laboratory studies often report a toxicological response to a specific dose of a contaminant, without measuring the body or organ concentration of the subject animals when the response occurs. Additionally, the dose is sometimes administered in a different

fashion (e.g., injection) than the animals would normally receive by environmental exposure.

Most of the field studies cited above concluded that the Pb levels found in mice were not present at toxic levels. In the study showing Pb accumulations of 52.1 mg/kg in bone tissue, the authors concluded that the level of Pb exposure required to produce recognizable poisoning in captive deer mice was about five times the level of exposure of mice in that study (62).

In contrast, another study<sup>(59)</sup> reported that the concentrations of Pb in deer mice in their study exceeded 10 mg/kg, a level of "diagnostic significance" in livers of experimental mammals. They further noted that mammals with behavioral and physiological signs of Pb intoxication have died with 5 mg/kg Pb or less in their livers.

### 4.4.2 Food Chain Effects

Regardless of the form in which it is encountered, about 2% of inorganic Pb which is ingested by mammals is absorbed<sup>(64)</sup>. Lead in the body tissue can be expected to be more available to consumers than the Pb which is present in the soil and sediment<sup>(51)</sup>. Snakes, raptors, and a variety of mammalian carnivores can be expected to accumulate Pb from consuming small mammals inhabiting the site.

Other species of small mammals may accumulate Pb to a greater extent than P. leucopus. Prior studies have found that shrews and voles accumulate higher amounts of Pb than Peromyscus (14,59,65). While shrews and other small mammal species may not be as abundant, they constitute components of the terrestrial food web.

### 5.0 CONCLUSIONS AND RECOMMENDATIONS

(1) Pb concentrations in West Stream sediment were highest near the NL facility. They were also higher in the West Stream than in the East Stream. Although sampling was limited, these results are consistent with the RI<sup>(1)</sup>.

- (3) Green frogs (Rana clamitans) had a mean Pb tissue concentration of 9 mg/kg wet weight in the West Stream (range 1.4 23 mg/kg). Tissue levels varied with sediment concentrations. The highest concentrations were noted at locations within the West Stream which were closest to the NL facility. The mean tissue level in the East Stream was lower (6 mg/kg). While these levels are probably not detrimental to frogs, they are minimum estimates since the frogs collected were nearly all subadults.
- (4) Earthworms (Eisenia foetida) exposed to a range of site soil Pb concentrations (120 to 6900 mg/kg) accumulated a maximum of 170 mg/kg tissue Pb (wet weight) over a 28-day exposure period. Tissue Pb concentrations were not correlated with soil Pb concentrations or other soil characteristics (grain size, TOC, percent moisture, pH) measured. Organic matter was significantly associated with Pb accumulation but had a poor correlation coefficient, and pH was close to neutral in most chambers. Prior studies have shown a strong influence of pH and organic matter on availability, suggesting that Pb in the soil is largely unavailable.
- (5) Bioconcentration factors (Pb in tissue divided by Pb in soil) decreased as soil Pb contamination increased. Thus, either Pb was less available at higher concentrations or worms exhibited aversion to feeding. These results are consistent with prior studies showing that worms accumulate a higher proportion of soil Pb at lower soil concentrations.
- (6) Pb levels found in the worms in this study could be detrimental to the worms, but other studies have reported higher levels of Pb accumulation. Worms in all chambers experienced significant weight loss which was weakly but significantly correlated with Pb (r=0.6). Lesions and other apparent abnormalities were noted in 8 out of the 200 worms tested. Mortality was noted in several chambers, but could be attributed to a variety of factors.
- (7) White-footed mice (*Peromyscus leucopus*) accumulated up to 13 mg/kg Pb (wet weight). This species is the most abundant and easily collected small mammal on site. However, based on results of prior studies, this species apparently does not

accumulate as much Pb as shrews and voles. These species are present on site, but were not as abundant, and hence would be more difficult to study.

- (8) Pb present in the tissue of earthworms, frogs, and mammals collected on site is present in a form more available to consumers higher in the food chain than Pb in the soil or sediment. However, Pb is not known to biomagnify in the food chain, and only a proportion of the available Pb should be assimilated. Resident species such as the red fox and greatblue heron would be expected to accumulate higher body burdens of Pb than species utilizing the site on a seasonal basis.
- (9) The forested wetlands in the vicinity of Grid IA and immediately north of grid III represent high quality habitat for breeding forest birds and resident mammals; this conclusion is based upon the best professional judgement of the investigators in this study.
- (10) Any sediment removal efforts should consider that disturbance of sediment could lower pH and potentially make Pb in other areas of the stream more available for uptake, regardless of concentration.
- (11) Soil removal efforts should consider the potential role of organic matter in immobilizing Pb. If trees are removed, a major source of organic material would be lost. It is conceivable that Pb in deforested areas would then be more available for uptake by terrestrial invertebrates.
- (12) Site soil characteristics should also be considered during removal. According to the U.S. Soil Conservation Service Salem County Soil Survey, on-site soils are highly subject to wind erosion, which would be expected to increase if large amounts of forest vegetation were removed.

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Table 1

In-Situ Water Quality Parameters at Selected Surface Water Locations
National Lead Site
Pedricktown, New Jersey
November, 1992

Sampling Location	Date	Time	Temp (°C)	pН	Dissolved Oxygen (mg/L)	Conduc- tivity (umhos/ cm)	Oxidation- Reduction Potential (volts)	Salinity (ppt)
East Stream -	6/17	1340	21.23	6.5	6.7	0.276	0.143	0.00
Pedricktown Road	8/10	1050	22.1	6.2	1.2	0.178	0.044	0.00
West Stream - Pedricktown Road	8/10	1110	23.3	5.9	4.9	0.276	0.143	0.00
West Stream -	6/10	1130	21.9	5.8	6.3	0.268	0.093	0.00
Route 130	6/10	1615	28.1	5.2	6.2	0.271	0.180	0.00
	6/17*	1400	27.2	6.0	6.9	0.330	0.183	0.00
	6/17	1420	29.0	6.0	6.3	0.332	0.150	0.00
	8/10	1155	25.5	6.2	9.8	0.307	0.062	0.00

Reading taken immediately north of the fence; remaining readings taken south of the fence.

Table 2
Summary of Pb Concentration Ranges in Sediment
National Lead
Pedricktown, New Jersey
November, 1992

Arca Investigated	Sample Locations <sup>(1)</sup>	Pb Concentration Range <sup>(2)</sup> (mg/kg)
West Stream South of Pedricktown Road	2,4,6	386-1100
West Stream Between Railroad Tracks and Pedricktown Road	8,11,13	4147-20,670
West Stream Immediately South of Route 130	10,12	939-1237
East Stream Near Goodrich Facility	14,16	36-72
East Stream Powerline, South of Railroad Tracks	1,5,7,15,17	53-1331
East Stream Woods North of Pedricktown Road	3,9	32-260

<sup>(1)</sup>Refer to Figure 3 for sediment sampling locations.

<sup>&</sup>lt;sup>CD</sup>Compiled from both XRF and AA data; AA data was recorded only at locations 2,4,5,8,and 9.

Table 3 Results of Pb, Field pH, Percent Moisture and TOC Analyses of Toxicity Testing Sediment Samples National Lead Site Pedricktown, New Jersey November, 1992

Sampling Location	Pb Concentration (mg/kg) <sup>(1)</sup>	Method Detection Limit (mg/kg)	Field pH <sup>(2)</sup>	Percent Moisture	Total Organic Carbon (g/kg)	Method Detection Limit (g/kg)
2 (West)	670	10	3.93	2	150.0	0.39
4 (West)	1100	10	6.09	3	85.0	0.36
5 (East)	53	10	6.08	2	4.8	0.16
8 (West)	4400	11	6.18	2	160.0	0.63
9 (East)	260	10	6.35	5	66.0	0.23

<sup>&</sup>lt;sup>(1)</sup>Measured by AA analysis.
<sup>(2)</sup>Field measured using an electronic pH meter.

Table 4
Grain Size of Sediment Samples
National Lead Site
Pedricktown, New Jersey
November, 1992

Sampling Location	% Granules <sup>(1)</sup>	% Sand <sup>ra</sup>	% Fine Sand <sup>(3)</sup>	% Silt/Clay(4)
2 (West)	0.00	8.55	14.80	76.64
4 (West)	4.43	9.08	37.60	48.89
5 (East)	1.93	68.81	23.92	4.67
8 (West)	0.00	18.84	37.50	43.66
9 (West)	0.34	40.26	44.45	14.94

<sup>(1) -</sup> Particles 2.0 mm and larger in size

<sup>&</sup>lt;sup>(2)</sup> - Particles between 0.30 mm and 2.0 mm in size

<sup>&</sup>lt;sup>(3)</sup> - Particles between 0.075 mm and 0.30 mm in size

<sup>(4) -</sup> Particles less than 0.075 mm in size

Table 6
Water Quality Parameters Measured from the Sediment Toxicity Test Water
National Lead Site
Pedricktown, New Jersey
November, 1992

Sampling Location	Dissolved Oxygen (mg/L)	Lab pH	Temperature (°C)	Conductivity (umhos/cm)	Total Alkalinity (mg/L)	Total Hardness (mg/L)
Control	5.6-8.4	6.4-7.0	20.0-23.5	160-195	16-20	56
2	7.0-8.7	3.7-4.1	20.0-23.5	230-260	<4	60-68
4	6.3-8.4	5.5-6.7	20.0-23.5	190-250	20-32	68
5	4.5-8.7	4.4-6.3	20.0-23.5	210-330	4-12	72-76
8	6.5-8.6	5.2-7.2	20.0-23.5	185-220	25-56	64-88
9	6.5-8.8	4.3-6.4	20.0-23.5	210-260	4-8	60-76

Table 7
Whole body (wet weight) Pb Concentrations and Metrics of Green Frogs (Rana clamitans)
National Lead Site
Pedricktown, New Jersey
November, 1992

Location Number	Pb Concentration (mg/kg)	Method Detection Limit (mg/kg)	Body Length (mm)	Body Weight* (g)	Percent Moisture
ES-1	13.0	0.50	60	15.988	78
ES-2	3.9	0.48	54	11.372	79
ES-3	3.4	0.45	41	5.098	83
ES-4	3.6	0.43	41	5.842	74
WS2A-1	17.0	0.43	35	3.366	81
WS2B-2	2.3	0.48	30	1.889	80
WS2C-3	23.0	0.40	40	4.696	82
WS2D-4	5.2	0.45	32	3.422	86
WS2E-5	9.4	0.41	33	3.230	81
WS2F-6	12.0	0.49	39	4.386	80
WS130-1	8.6	0.37	40	5.354	82
WS130-2	1.4	0.50	53	13.586	84
WSP-1	3.9	0.46	34	2.855	79

<sup>&</sup>quot;Wet weight taken after stomach contents and intestines were removed.

Table 8
Pb Concentrations in Sediment at Green Frog (Rana clamitans) Capture Points
National Lead Site
Pedricktown, New Jersey
November, 1992

Location Number <sup>(1)</sup>	Frog Pb Concentration (mg/kg)	Location Description <sup>(1)</sup>	Approximate Distance to XRF Sample Point (ft)	XRF Location Number	Sediment Pb Concentration <sup>(2)</sup> (mg/kg)
ES-1	13.0	East Stream,	10-50	ES-FS	782
ES-2	3.9	Pedricktown Road			549°
ES-3	3.4				
ES-4	3.6				
Mean Pb Value	5.98				
WS130-1	8.6	West Stream,	10-30	130-FS	835
WS130-2	1.4	Route 130			886 <sup></sup> 1352 <sup></sup>
Mean Pb Value	5.0				
WS2A-1	17.0	West Stream, South of	10-25	WS-FS	4524
WS2C-3	23.0	Railroad Tracks			4611°
WS2D-4	5.2				
WS2E-5	9.4				
WS2F-6	12.0				
Mean Pb Value	13.32				
WSP-1	3.9	West Stream,	40	#4	1041
WS2B-2	2.3	Pedricktown Road			994 <b>-</b> 946 <b>-</b>
Mena Pb Value	3.1				

Duplicate reading from same sample.

Replicate reading from same sample (different aliquot).

<sup>(1)</sup>Refer to Figure 4.

<sup>&</sup>lt;sup>CD</sup>As measured by XRF.

Table 9
Soil Lead Concentrations at Earthworm Chamber Locations
National Lead Site
Pedricktown, New Jersey
November 1992.

Chamber Location #	XRF Location	XRF Surface PB Concentration (mg/kg)	XRF Homogenate Pb Concentration (mg/kg)	AA Homogenate Pb Concentration
1	E16	2819	313	290
2	E2	1001	216	190
3	El	3041	1035	810
4	E11	211	169	180
5	E17	2015	1351	1100
6	E18	1501	859	720
7	E4	888	435	450
8	W1	3937	2941	1800
9	W12	2846	5371	3500
10	₩3	2518	1234	830
11	<b>W</b> 5	1746	1446	1300
12	₩9	3088	1737	1600
13	₩8	1885	2089	1500
14	W14	4344	4322	2200
15	<b>W</b> 16	2706	3640	1800
16	W18	3184	9401	6700
17	W19	9965	8967	6800
18	W20	12737	10082	6900
19	W22	5394	2571	2600
20	E28	350	. 241	120

Table 10 Soil pH, Moisture, and Total Organic Content (TOC) at Earthworm Chamber Locations National Lead Site Pedricktown, New Jersey November 1992.

Chamber Location #	pH Value	Percent Moisture <sup>(1)</sup> (Field)	Percent Moisture (Laboratory)	TOC Concentration (g/kg) <sup>(2)</sup>
Eastern Locations				
1	7.0	10	16.09	26
2	7.0	10	11.16	13
3	7.0	5	20.14	64
4	7.0	10	11.66	17
5	7.0	12	22.08	68
6	7.0	10	18.99	55
7	7.0	20	19.55	31
20	7.0	55	66.04	210
Western Locations				
8	6.9	50	65.62	180
9	6.2	68	53.40	140
10	7.0	20	50.05	120
11	7.0	15	34.23	99
12	7.0	5	30.60	90
13	6.9	30	28.16	120
14	6.9	21	61.82	200
15	7.0	40	64.12	190
16	6.8	40	36.44	41
17	6.6	57	69.14	170
18	7.0	60	63.15	190
19	6.7	50	58.31	130

<sup>&</sup>lt;sup>(1)</sup>Measured by a hand held soil moisture meter. <sup>(2)</sup>All concentrations based upon dry weight.

Table 19

Mean Whole Body Wet Weight Pb Concentrations in Peromyscus leucopus

National Lead Site

Pedricktown, New Jersey

November, 1992

Area	Number of Animals	Mean Pb Concentration <sup>(1)</sup> (mg/kg)	Standard Deviation	Range of Values (mg/kg)
	11	1.60	1.10	0.2-3.3
	15	3.10	3.02	0.9-13
	12	4.77	3.50	0.9-13
James	38	3.16	2.54	0.2-13

<sup>&</sup>lt;sup>(1)</sup> Untransformed means; includes adults, subadults, and juveniles.

Table 12

Percent Organic Matter of Soil Samples Collected from Earthworm Chambers
National Lead Site
Pedricktown, New Jersey
November, 1992

Sample Location #	Percent Organic Matter <sup>(1)</sup>				
Eastern Locations					
1	7.96				
2	4.20				
3	20.74				
4	5.82				
5	21.90				
6	18.34				
7	12.56				
Western Locations					
20	53.66				
8	54.99				
9	59.56				
10	47.88				
11	34.47				
12	30.86				
13	31.05				
14	75.92				
15	78.26				
16	17.87				
17	48.75				
18	48.54				
19	34.03				

 $<sup>^{(1)}</sup>$ As measured by percent of sample lost upon ignition at 700° F; see Appendix C.

Table 13

Earthworm Survival, Lead Concentrations and Percent Moisture
National Lead Site
Pedricktown, New Jersey
November, 1992

Chamber Location #	Soil Pb Concentration (mg/kg) <sup>(1)</sup>	Survival (# Worms/ Chamber) <sup>(2)</sup>	Earthworm Po Concentration (mg/kg Wet Weight)	Earthworm Percent Moisture
1	290	10	58	84
2	190	8	48	84
3	810	8	130	85
4	180	10	29	86
5	1100	9	52	84
6	720	7	34	84
7	450	7	130	85
8	1800	8	71	82
9	3500	7	100	81
10	830	9	76	85
11	1300	0	*	*
12	1600	0	<b>#</b> c	•
13	1500	10	46	85
14	2200	7	170	81
15	1800	9	63	85
16	6700	3	62	82
17	6800	7	67	84
18	6900	2	140	84
19	2600	0	•	•
20	120	3	*	•
Lab Reference	NR	NA	ND	84
4 (10-day rep)	180	9	15	86
4 (20-day rep)	180	10	21	85
4 (20-day dup)	180	10	19	85

<sup>(1)</sup>AA values

<sup>&</sup>lt;sup>∞</sup>10 worms

<sup>+ -</sup> not analyzed due to mortality

ND - not detected above Method Detection Limit of 0.25 mg/kg

NR - not recorded

NA - not applicable; not exposed for 30 days

Table 14

Earthworm Lead Bioconcentration Factors<sup>(1)</sup>

National Lead Site

Pedricktown, N.J.

November, 1992

Soil Pb Concentration Range (mg/kg)	Number of Chambers	Mean Concentration Factor (CF)	CF Range
< 500	4	0.23	0.16-0.29
500-1000	3	0.10	0.05-0.16
1000-2000	3	0.04	0.03-0.05
2200	1	0.08	NA
3500	1	0.03	NA
>6500	3	0.01	0.01

<sup>(1)</sup> Defined as wet weight Pb concentration in worm tissue divided by soil Pb concentration.

\*Concentration Factors could not be calculated for chambers 11,12,19 and 20 due to low survival.

NA - Not Applicable.

1 .

Table 15
Mean Individual Earthworm Weights<sup>(1)</sup>
National Lead Site
Pedricktown, New Jersey
November, 1992

Chamber	Soil Pb	Initial	Weight	(g)	Fina	l Weight	(g)
Number	(mg/kg)	x	n	Ś	x	n	S
1	290	.562	10	.080	.363	10	.084
2	190	.496	10	.082	.321	8	.053
3	810	.599	10	.074	.375	8	.075
4	180	.545	10	.080	.355	10	.073
5	1100	.538	10	.069	.367	9	.065
6	720	.563	10	.085	.342	7	.079
7	450	.458	10	.112	.349	7	.056
8	1800	.515	10	.086	.290	8	.038
9	3500	.670	10	.086	.312	9	.034
10	830	.460	10	.086	.300	9	.063
11	1300	.471	10	.061	*	*	
12	1600	.489	10	.055	*	*	*
13	1500	.440	10	.088	.320	10	.083
14	2200	.592	10	.077	.328	7	.034
15	1800	.584	10	.112	.359	9	.043
16	6700	.552	10	.116	.289	3	.130
17	6800	.557	10	.154	.268	7	.077
18	6900	.573	10	.155	. 155	2	.019
19	2600	.475	10	.077	*	*	•
20	120	.520	10	.144	.221	3	.091
10-Day Rep.	180	.502	10	.117	.475	9	.101
20-Day Rep.	180	.464	10	.051	**	**	**

<sup>(1)</sup> All weights are wet weights

<sup>\*</sup> Indicates no worms were found

<sup>\*\*</sup>Indicates worms were not weighed

## Table 16 Surface Pb Concentrations(1) at Small Mammal Trapping Grids National Lead Site Pedricktown, New Jersey November, 1992

Area Location	XRF Surface Location Numbers <sup>(2)</sup>	Median Pb Concentration (mg/kg)	Mean Pb Concentration (mg/kg) <sup>G,4</sup>	Range of Values <sup>(5)</sup> (mg/kg)
Grid 1	W1-W11	1776	1953	148-4177
Grid IA	N1-N15	1450	1515	287-2378
Area I (Grids I and IA)	above	1650	1627	148-4177
Area II	E1-E21	640	940	171-2819
Area III	E26-E28, EE1-EE10	2031	2286	438-4930

<sup>&</sup>lt;sup>(1)</sup>XRF

<sup>&</sup>lt;sup>CD</sup>Refer to Figure 5 for geographic locations.

<sup>&</sup>lt;sup>(5)</sup>Means calculated do not include replicate readings.

<sup>&</sup>lt;sup>(4)</sup>Statistical comparisons between Grid Areas were not made since data were not randomly collected.

<sup>&</sup>lt;sup>(5)</sup>A complete list of values is given in Appendix A, XRF Results.

Table 17
Small Mammal Trap Success
National Lead Site
Pedricktown, New Jersey
November 1992.

Species	Grid I	Grid IA	Grid II	Grid III	Total
Peromyscus leucopus	4	9	15	12	38
Pitimys pinetorum	0	0	12	1	12
Blarina brevicauda	2	0	1	1	4
TOTALS	6	9	28	14	54
Number of trap-nights	300	100	450	300	1150
Percent success	0.02	9.0	6.0	5.0	4.7

Table 18
Body Metrics of Adult Peromyscus leucopus<sup>(1)</sup>
National Lead Site
Pedricktown, New Jersey
November, 1992

Area	n	Body we	eight (g) s	Liver W X	eight (g) s	Kidney W	eight <sup>(2)</sup> (g)
I	10	22.05	3.02	1.33	0.31	0.35	0.14
П	13	22.14	4.53	1.29	0.40	0.38	0.08
Ш	11	22.70	3.53	1.30	0.29	0.37	0.08
Overali	34	22.30	3.69	1.31	0.33	0.37	0.1

Area	n			Thymus	Thymus Weight (g)		Adrenal <sup>(3)</sup> Weight (g)	
		x	g) s	x	S	x	s	
I	10	0.064	0.031	0.006	0.00843	0.0096	0.00662	
П	13	0.062	0.032	0.008	0.00924	0.0074	0.00481	
ш	11	0.053	0.020	0.007	0.00815	0.0078	0.00349	
Overall	34	0.060	0.028	0.007	0.00861	0.0083	0.00497	

Area	Total Number of Animals	Number of Females	5	Weight g)	Number of Males	Wei (	Testes ght <sup>(4)</sup> g)
	<del></del>			8		X	<b>s</b>
I	10	6	0.198	0.288	4	0.3335	0.2793
11	13	5	0.091	0.091	8	0.4764	0.2390
Ш	11	4	0.005	0.010	7	0.4916	0.1261
Overall	34	15	0.111	0.197	19	0.4338	0.6444

 $<sup>^{0}</sup>$ No statistically significant differences were noted between areas.

<sup>&</sup>lt;sup>Co</sup>Value represents combined weight of both kidneys.

OValue represents combined weight of both adrenals.

<sup>&</sup>lt;sup>64</sup>Value represents combined weight of both testes.

X, s and n refer to mean, standard deviation and number of individuals, respectively.

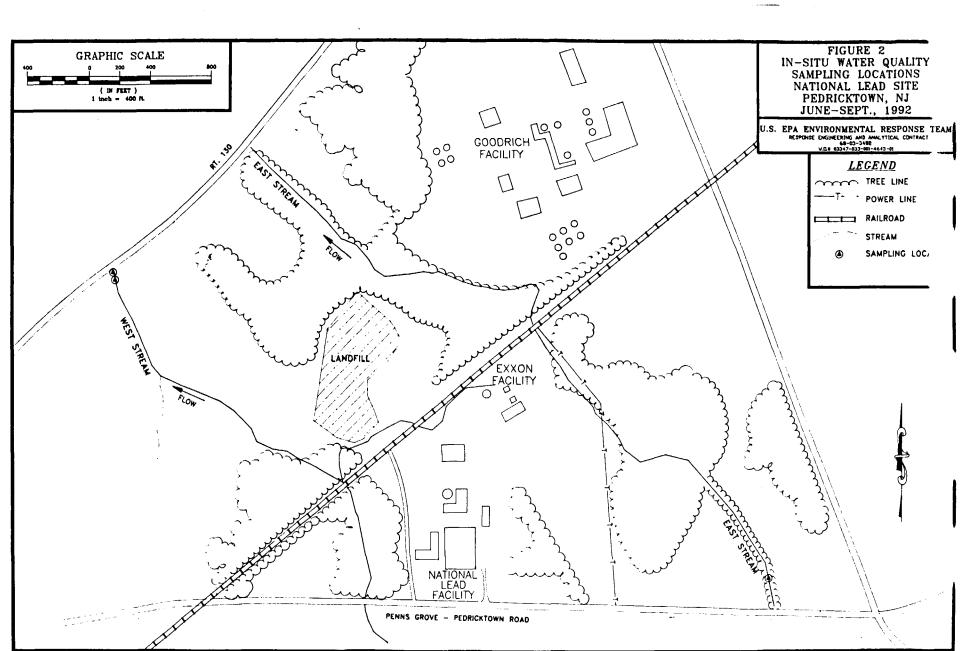
Table 19

Mean Whole Body Wet Weight Pb Concentrations in Peromyscus leucopus
National Lead Site
Pedricktown, New Jersey
November, 1992

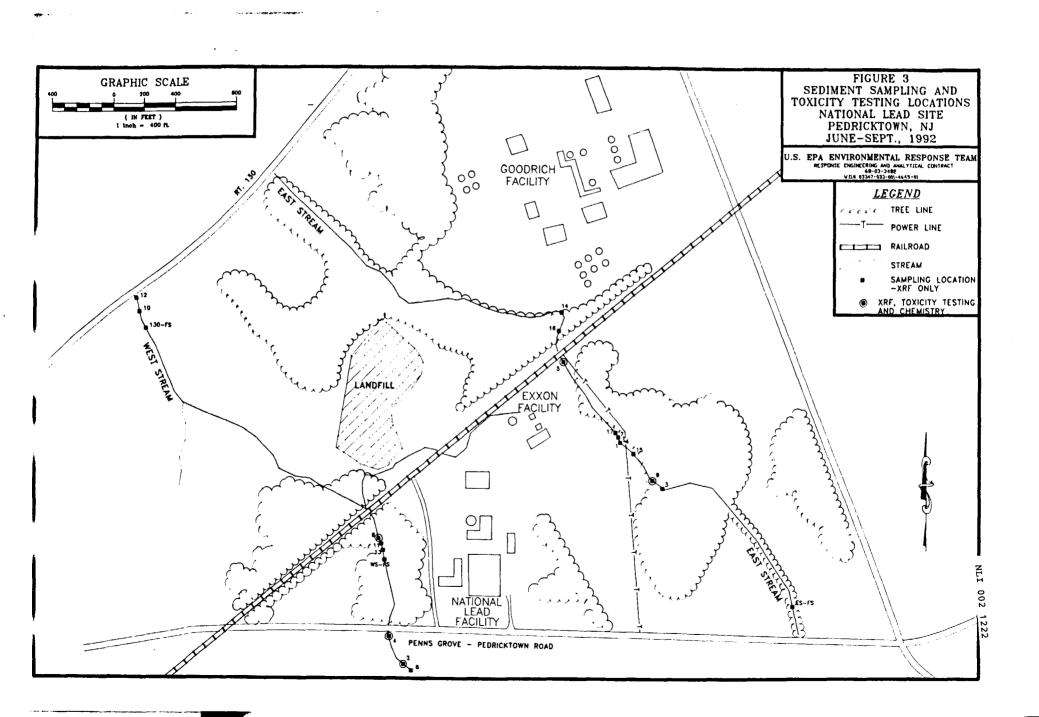
Area	Number of Animals	Mean Pb Concentration(1) (mg/kg)	Standard Deviation	Range of Values (mg/kg)
I,IA	11	1.60	1.10	0.2-3.3
П	15	3.10	3.02	0.9-13
Ш	12	4.77	3.50	0.9-13
Overall	38	3.16	2.54	0.2-13

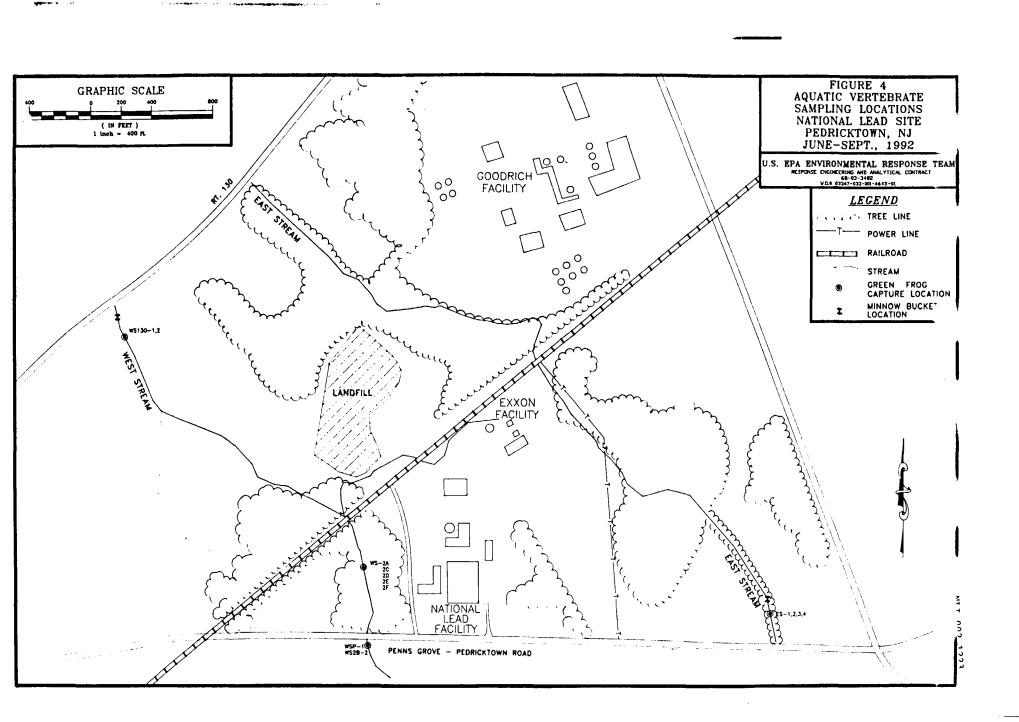
<sup>(1)</sup> Untransformed means; includes adults, subadults, and juveniles.

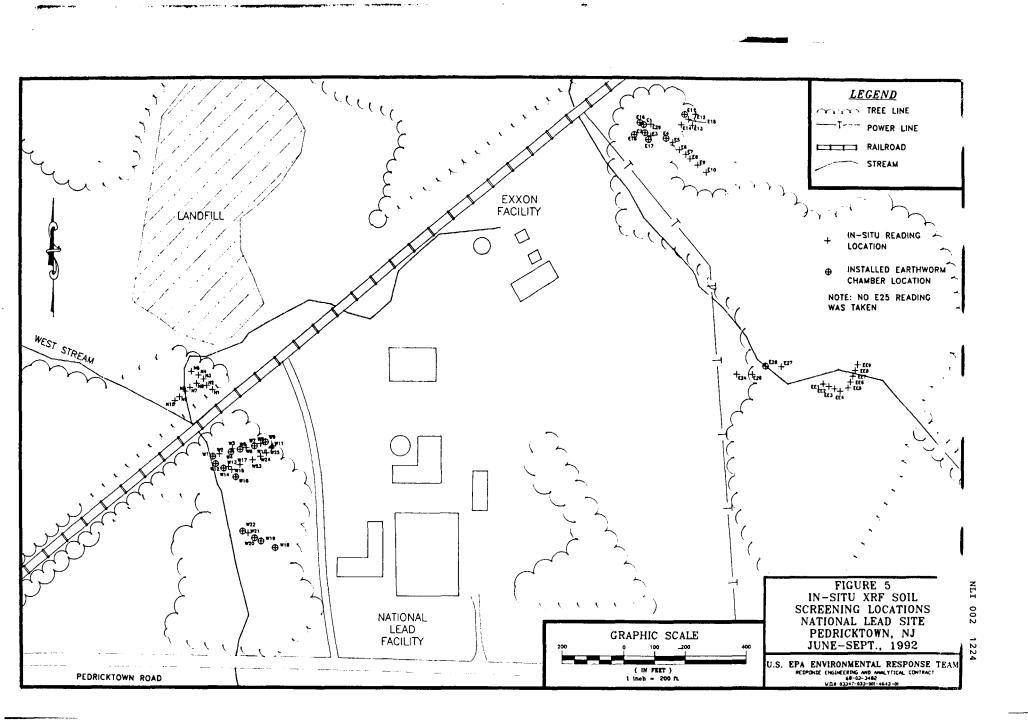
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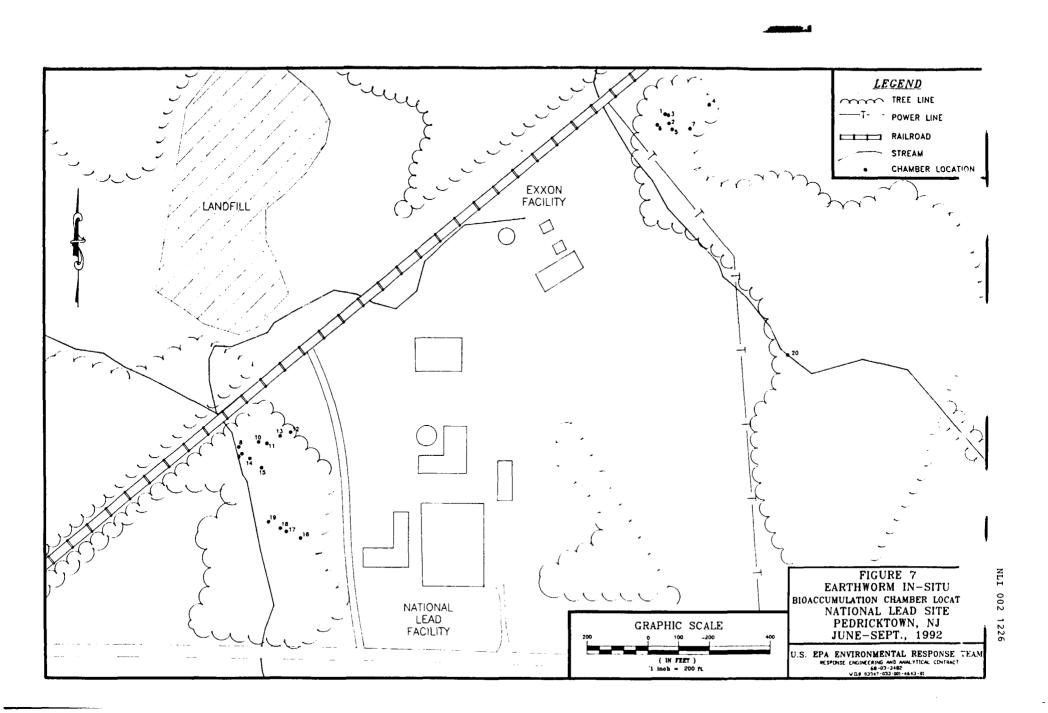


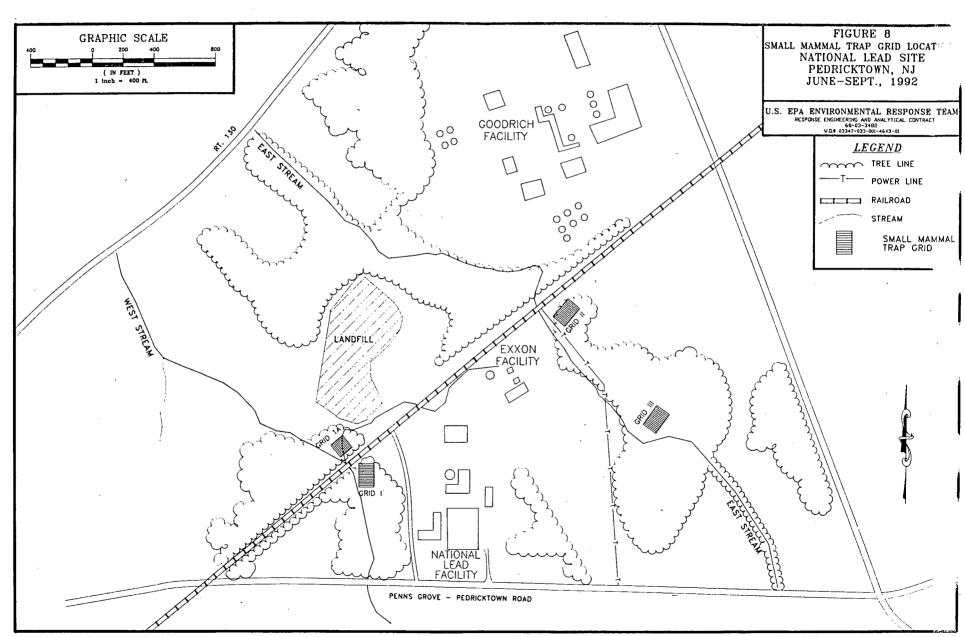
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